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(54) Title: CHIMERAS OF HEPATITIS C VIRUS AND BOVINE VIRAL DIARRHEA VIRUS

(57) Abstract

Disclosed is a polynucleotide comprising a chimeric viral RNA which contains: a 5' nontranslated region (5' NTR), an open reading frame (ORF) region, and a 3' nontranslated region (3' NTR) wherein at least one of said regions is chimeric. The chimeric region comprises a first nucleotide sequence from a pestivirus in operable linkage with a heterologous nucleotide sequence. The chimeric viral RNA is replication-competent. Preferably the pestivirus sequence is from a bovine viral diarrhea virus and the heterologous nucleotide sequence is from a hepatitis C virus. Also disclosed are a method for identifying compounds having antiviral activity agaisnt hepatitis C virus, a genetically-engineered chimeric RNA virus and a vaccine against bovine viral diarrhea virus.

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Chimeras of Hepatitis C Virus and Bovine Viral Diarrhea Virus

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Related Applications

This application claims priority to, and incorporates herein in its entirety, U.S. 60/082,964 filed April 24, 1998.

Background of the Invention

(1) Field of the Invention

This invention relates generally to the development of therapies for treating hepatitis C virus (HCV) and bovine viral diarrhea virus (BVDV) and more particularly to the identification of such therapies using chimeric viruses comprising a genomic sequence derived from HCV and bovine viral diarrhea virus (BVDV).

(2) Description of the Related Art

The *Flavivirdae* is an important family of human and animal RNA viral pathogens (Rice, CM. 1996. *Flavivirdae*: The viruses and their replication. In: Fields BN, Knipe DM, Howley PM., eds. *Fields virology.* Philadelphia: Lippincott-Raven Publishers. pp. 931-960.) The three currently recognized genera of the *Flavivirdae* family exhibit distinct differences in transmission, host range, and pathogenesis. For example, members of the classical flavivirus genus, such as yellow fever virus and dengue virus, are typically transmitted to vertebrate hosts via arthropod vectors and cause acute self-limiting disease (Monath TP, Heinz FX. 1996. Flaviviruses. In: Fields BN, Knipe DM, Howley PM., eds. *Fields virology.* New York: Raven Press. pp. 961-1034). The pestiviruses, such as bovine viral diarrhea virus (BVDV) and classical swine fever virus (CSFV), cause economically important livestock disease and are spread by direct contact or the fecal-oral route (Thiel et al., 1996. Pestiviruses. In: Fields BN, Knipe DM, Howley PM., eds. *Fields virology.* New York: Raven Press. pp. 1059-1073). The most recently characterized *Flavivirdae* genus is the hepacivirus genus, the sole member of which is the common and exclusively human pathogen, hepatitis C virus (HCV). HCV is

transmitted by contaminated blood or blood products and is the most common agent of non-A, non-B hepatitis, affecting more that 1% of the population worldwide (Houghton, 1996. Hepatitis C viruses. In: Fields BN, Knipe DM, Howley PM., eds. *Fields virology*. Philadelphia: Lippincott-Raven Publishers. pp. 1035-1058.). Unlike flavivirus and pestivirus infections, which are usually eliminated by host immune response, chronic HCV infections are common and can cause mild to severe liver disease including cancer.

Despite these differences, members of the *Flavivirdae* family share common structural features and gene expression strategies. Virus particles consist of a lipid bilayer envelope with embedded transmembrane glycoproteins surrounding a protein-RNA nucleocapsid. Genome RNAs are single-stranded of positive polarity, and function as the sole mRNA species for translation of a single long open reading frame (ORF). This ORF is translated into a polyprotein which is processed by cellular and viral proteases into mature viral proteins. Structural proteins destined for incorporation into virus particles are encoded in the N-terminal portion of the polyprotein, while the nonstructural proteins which form components of the viral RNA replicase are encoded in the remainder.

Replication of the *Flavivirdae* RNA genome occurs via synthesis of a full-length negative-strand intermediate and is asymmetric, favoring synthesis of positive-strand RNAs. However, little is known about the details of this process. For all three genera of the *Flavivirdae* family, full-length functional cDNA clones have been constructed and RNAs transcribed from these cDNA templates are infectious. For flaviviruses and pestiviruses, mutagenesis of these clones and efficient RNA transfection of permissive cell cultures provides a means of probing the role of *cis* RNA elements and viral proteins in replicase assembly and function. Such analyses are not yet possible for HCV since this virus is unable to replicate efficiently in cell culture.

Like many other RNA viruses, it is believed the 5' and 3' terminal sequences of the *Flavivirdae* contain conserved *cis*-elements important for translation, RNA replication, and packaging (Bukh et al., *Proc. Natl. Acad. Sci. USA 89*:4942-4946, 1992; Deng et al., *Nucleic Acids Res. 21*:1949-1957, 1993; Cahour et al., *Virol. 207*:68-76, 1995; Kolykhalov et al., *J. Virol. 70*:3363-3371, 1996; Men et al., *J. Virol. 70*:3930-3937, 1996; Tanaka et al., *J. Virol. 70*:3307-3312, 1996; Huang HV. 1997. Evolution of the alphavirus promoter and the *cis*-acting sequences of RNA viruses. In: Saluzzo J-F, Dodet B. eds. *Factors in the emergence of arbovirus disesases*. Paris: Elsevier Press, pp. 65-79; Mandl et al., *J. Virol. 72*:2132-2140, 1998). The 5' nontranslated region (NTR) functions initially at the level of translation. Similar to most cellular mRNAs, flavivirus genome RNAs are translated in a cap-dependent manner. These RNAs contain a 5' cap structure that is presumably added by virus-encoded

RNA triphosphatases, guanylyl-, and methyl-transferases (Rice, 1996, *supra*). In contrast, the translational strategy employed by pestiviruses and HCV is more similar to that of the picornaviruses. These RNAs appear to be uncapped and contain long 5' NTRs with *cis* RNA elements that function as internal ribosome entry sites (IRES) for translation initiation at the polyprotein AUG (Lemon et al., *Semin. Virol. 8*:274-288, 1997).

The 5' NTRs of HCV and BVDV have a similar structural and functional organization despite containing only short stretches of high sequence identity (Wang et al., *Curr. Top. Microbiol Immunol. 203*:99-115, 1995; Lemon et al., 1997, *supra*). The IRES within each NTR is located at the 3' end of the NTR at a position proximal to the AUG initiation codon of the ORF. Although the 5' terminal sequence of each of these viruses is apparently not required for IRES function (Rijnbrand et al., *FEBS Lett* 365:115-119, 1995; Honda et al., *Virology. 222*:31-42, 1996; Rijnbrand et al., *J. Virol. 71*:451-457, 1997), these sequences are highly conserved among different strains of HCV (Bukh et al., *Proc. Natl. Acad. Sci. USA:89*:4942-4946, 1992) or BVDV (Deng et al., 1993, *supra*), suggesting they play other roles in viral replication. For example, sequences in the 5' NTR may be required for regulating translation versus initiation of negative-strand RNA synthesis. Such regulation could occur by direct interaction of 5' and 3' RNA elements or indirectly, via RNA-protein interactions. Sequences in the 5' NTR may also modulate packaging versus translation. Finally, sequences complementary to the 5' NTR, which are located at the 3' end of negative-strand RNA, are likely to function in the initiation of positive-strand RNA synthesis.

The HCV 3′ NTR contains an internal polypyrimidine tract followed by a highly conserved sequence of 98 bases at the 3′ terminus, which has been shown to be required for replication of HCV (U.S. Application Serial No. 08/811,566).

Further elucidation of the role of sequences in the HCV 5′ and 3′ NTRs has been hampered by the inefficient replication of HCV in cell culture. This aspect of HCV biology also makes it difficult to identify and test possible antiviral compounds for activity against HCV. Thus, a need exists for a system which facilitates investigation of HCV replication and therapeutic approaches to control HCV infections.

Summary of the Invention

Briefly, therefore, the present invention provides novel compositions and methods for studying HCV replication which are based on the discovery that chimeras of HCV and BVDV genomic sequences can be constructed that are able to replicate in cell culture. The BVDV-specific sequence provides the chimeric viral nucleic acid with the ability to replicate in cell culture, while the HCV-specific sequence allows the chimeric viral nucleic acid to be used to

screen possible compounds for anti-viral activity against HCV. It is believed that similar replication-competent chimeras can be constructed from HCV and other pestiviruses.

Thus, in one embodiment, the present invention provides a novel, chimeric viral RNA in which at least one of the 5′ NTR; ORF and 3′ NTR regions is chimeric and comprises a nucleotide sequence from the corresponding region of a pestivirus in operable linkage with a nucleotide sequence from the corresponding region of an hepatitis C virus (HCV). The chimeric viral RNA is replication-competent. In preferred embodiments, the pestivirus is BVDV.

In other embodiments, the invention provides a polynucleotide comprising a DNA-dependent promoter operably linked to a cDNA of a chimeric viral RNA as described above and cells transiently transfected or stably transformed with the polynucleotide. In some embodiments the cDNA may encode a dominant selectable marker or an assayable reporter.

In yet another embodiment, the invention provides a method for identifying compounds having anti-HCV activity. The method comprises providing a first cell containing a chimeric viral nucleic acid derived from HCV and a pestivirus as described above and a second cell containing the pestivirus, and then comparing the replication efficiency of the chimeric viral nucleic acid in the presence and absence of a test compound to the replication efficiency of the pestivirus in the presence and absence of the test compound, wherein a greater reduction in compound-induced replication efficiency of the chimeric viral nucleic acid than the pestivirus indicates the compound has anti-HCV activity.

The invention also provides a genetically-engineered virus which comprises a chimeric viral nucleic acid derived from HCV and a pestivirus as described above. In one embodiment the genetically-engineered virus comprises virus particles containing at least one HCV structural protein and is useful in a vaccine against HCV. In another embodiment, the genetically-engineered virus is attenuated as compared to the pestivirus and is useful as a vaccine against the pestivirus.

In a still further embodiment, the invention provides a replication-competent BVDV vector expressing a heterologous sequence. The BVDV vector comprises the BVDV sequences encoding the BVDV replication machinery. In some embodiments, the replication-competent BVDV vector expresses an antigen and is useful as a vaccine.

Brief Description of the Drawings

Figure 1 is a schematic representation of the 5' NTRs of BVDV, HCV, and EMCV showing the position of the start codons of the ORF, and the boxes indicating the canonical IRES elements.

Figure 2 shows a schematic representation of BVDV and HCV chimeras, plaque phenotypes, reticulocyte translation efficiencies relative to parental BVDV, specific infectivities in MDBK cells, titers at 24 and 48 h post-transfection (or 72 h, as indicated), and an indication of whether pseudorevertants arose with results from BVDV, 5'HCV, BVDV+HCV, and BVDV+HCVdelB3 chimeras shown in Fig. 2A and results from BVDV+HCVdelB2B3, BVDV+HCVdelB1B2B3, BVDV+HCVdelB2B3H1, and BVDV+HCVdelB2B3H1H2 shown in Fig. 2B, where N.D. means not determined.

Figure 3 illustrates the *in vitro* translation efficiency of BVDV RNA or chimeras showing bar graphs of the amount of N^{pro}, the N-terminal protein in the BVDV ORF, expressed by the various constructs.

Figure 4 illustrates a schematic representation of EMCV chimeras, plaque phenotypes, reticulocyte translation efficiencies relative to parental BVDV, specific infectivities in MDBK cells, titers at 24 and 48 h post-transfection (or 72 h, as indicated), and an indication of whether pseudorevertants arose.

Figure 5 illustrates a pseudorevertant analyses showing in (Fig. 5A) the relative positions of mutations detected within the plaque-purified variants of passaged BVDV+HCVdelB1B2B3, 5'EMCV, and 5'HCV, and in (Fig. 5B) the 5' terminal sequences of pseudorevertants of BVDV+HCVdelB1B2B3, 5'EMCV, and 5'HCV. Novel nucleotides or sequences are shown in bold upper case type. Pseudorevertants are numbered and designated by the suffix ".R". The upper case sequence in BVDV+HCVdelB1B2B3 and BVDV+HCVdelB1B2B3.R1 is a remnant of downstream BVDV 5' NTR sequences and was created during the cloning procedures.

Figure 6 illustrates the construction of derivatives of 5'HCV designed to contain 5' termini corresponding to the sequence detected within the three analyzed pseudorevertants. Fig. 6A shows the 5' terminal sequence of the 5'HCV derivatives with the suffix (orig) designating a derivative containing the original 5' terminal sequence of the pseudorevertant; the suffix (cons) designating a derivative containing the consensus tetranucleotide sequence 5'-GUAU at the same position; and novel sequences shown in bold upper case type. Fig. 6B shows plaque phenotypes, reticulocyte translation efficiencies relative to parental BVDV, specific infectivities in MDBK cells, and titers at 24 and 48 h post-transfection are indicated.

Figure 7 illustrates a single step growth curve for various chimeric constructs showing released virus titers measured by performing plaque assays on MDBK cells transfected with various constructs.

Figure 8 illustrates replication of BVDV RNA or chimeric derivatives in transfected MDBK cells. Equal numbers of MDBK cells (~ 8 x 10^6) were electroporated with 5 □g of

each *in vitro* synthesized RNA. MDBK cells were also transfected with infectious yellow fever 17D and Sindbis RNAs to provide molecular mass markers. One fifth of the transfected cells were seeded on 35-mm dishes and incubated in D-MEM supplemented with 10% horse serum for 6 h at 37°C. The media were then replaced with 1 ml of fresh media containing 2 g/ml of actinomycin D and 40 Ci/ml of ^{3}H-uridine. Incubations were continued for 10 h at 37°C. RNAs were isolated as described in Materials and Methods, and 1/4 of the samples was denatured in glyoxal and loaded on an agarose gel. (A) Autoradiograph of the dried gel. Only the portion of the gel containing the genomic RNAs is shown. (B) Amount of radioactivity contained within the displayed fragments as determined by scintillation counting. BVDV, lane 1; 5'HCV, lane 2; BVDV+HCVdelB2B3, lane 3; BVDV+HCVdelB2B3H1, lane 4; 5'HCV.R1orig, lane 5; 5'HCV.R1cons, lane 6; 5'HCV.R3orig, lane 7; 5'HCV.R3cons, lane 8; 5'HCV.R2orig, lane 9; 5'HCV.R2cons, lane 10; yellow fever 17D, lane 11; Sindbis, lane 12; non-transfected MDBK cells, lane 13. The experiments shown is one of two repetitions which yielded similar results.

Figure 9 illustrates the genetic map of plasmid pACNR/BUD.

Figure 10 illustrates the sequence of low copy number plasmid pACNR/BVDV NADL (circular) harboring the functional cDNA of cytopathic BVDV NADL (positive sense cDNA 5' to 3'; nt 1-12578.

Figure 11 illustrates the sequence of infectious BVDV NADL (positive sense cDNA 5' to 3').

Figure 12 illustrates the sequence of infectious non-cytopathic BVDV NADL lacking cIns (positive sense cDNA 5' to 3').

Figure 13 illustrates the sequence adapted HCV 5' NTR from 5'HCV/R1.cons (positive sense cDNA 5' to 3'; only the sequence from the 5' base to the ATG initiating the polyprotein is shown).

Figure 14 illustrates the sequence of adapted HCV 5' NTR from 5'HCV/R1.orig (positive sense cDNA 5' to 3'; only the sequence from the 5' base to the ATG initiating the polyprotein is shown).

Figure 15 illustrates the sequence of adapted HCV 5'NTR from 5'HCV/R2.cons (positive sense cDNA 5' to 3'; only the sequence from the 5' base to the ATG initiating the polyprotein is shown).

Figure 16 illustrates the sequence of adapted HCV 5' NTR from 5'HCV/R2.orig (positive sense cNDA 5' to 3'; only the sequence from the 5' base to the ATG initiating the polyprotein is shown).

Figure 17 illustrates the sequence of adapted HCV 5' NTR from 5'HCV/R3.cons (positive sense cDNA 5' to 3'; only the sequence from the 5'base to the ATG initiating the polyprotein is shown).

Figure 18 illustrates the sequence of adapted HCV 5'NTR from 5'HCV/R3.orig (positive sense cDNA 5' to 3'; only the sequence from the 5' base to the ATG initiating the polyprotein is shown).

Figure 19 illustrates the sequence of prototype HCV-BVDV chimera from pNADL/5'HR3.orig/3'H3'B with the adapted HCV 5'NTR from 5'HCV/R3.orig and tandem 3' NTR elements from HCV followed by BVDV (positive sense cDNA 5' to 3') as discussed in Example 5.

Figure 20 illustrates various deletions of the poly U track in the 3'NTR HCV sequence of BVDV/HCV chimera p5H-3H33.

Figure 21illustrates the schematic representation of functional HCV/-BVDV chimera from pCBV/p7.

Figure 22 illustrates the sequence of functional HCV-BVDV chimera from pCBV/p7 (positive sense cDNA 5' to 3').

Figure 23 illustrates the schematic representation of a HCV/BVDV chimera with selectable marker.

Figure 24 illustrates the sequence of functional HCV-BVDV chimera from pCBV/p7/IRES-pac expressing a dominant selectable marker conferring resistance to puromycin (positive sense cDNA 5' to 3').

Figure 25 illustrates the schematic representation of a bicistronic HCV/BVDV chimera.

Figure 26 illustrates the sequence of functional bicistronic chimera expressing the entire HCV structural region derived from plasmid pNADL/BI#41/HCV str (positive sense cDNA 5' to 3')

Description of the Preferred Embodiments

In accordance with the present invention, the inventors herein have succeeded in generating HCV-BVDV chimeric RNAs which are replication competent. Such chimeras are useful in screening compounds *in vitro* for antiviral activity against HCV. In addition, it is believed that *in vivo* replication of HCV-BVDV chimeras according to the invention may be attenuated as compared to wild-type BVDV and thus may be useful in vaccinating animals against BVDV. It is also believed that the HCV chimeric structures described herein for BVDV are applicable to other pestiviruses.

In the context of this disclosure, the following terms will be defined as follows unless otherwise indicated:

"Cis-acting sequences" means the nucleotide sequences from an RNA virus genome that are necessary for recognition of the genomic RNA by specific protein(s) of the RNA virus or host cell that carry out replication, transcription, translation or packaging of the genome.

"Genetically-engineered virus" means any virus whose genome is different than that of a wild-type virus due to a human-made deletion, insertion, or substitution of one or more nucleotides to the wild-type viral genome.

"Infectious" when used to describe a virus means the virus is capable of entering cells and initiating a virus replication cycle, whether or not this leads to the production of new RNA virus particles.

"Nucleotide sequence" as used herein refers to DNA and the corresponding RNA sequence where relevant. It will be understood that sequences shown in the Figures are DNA versions of the RNA sequence and that chimeric molecules of the invention may comprises RNA molecules or cDNA copies of such RNA molecules.

"Replication-competent" as applied to a chimeric HCV-pestivirus RNA means the RNA is capable of RNA-dependent replication in at least one cell type that supports replication of the wild-type parental pestivirus. The number of replicated RNA molecules produced by an HCV-pestivirus chimeric RNA of the invention is at least 10-fold higher than the limit of detection, which is typically 10 to 100 molecules. More preferably, chimeric RNA production by the HCV-pestivirus chimeric RNA is at least 10^2 to 10^3-fold higher than the detection limit. The replication-competent chimeric RNA replicates at an efficiency that is preferably, at least 0.001%, more preferably, at least 0.01%, more preferably, at least 0.1%, more preferably, at least 1%, more preferably at least 10% and most preferably at least 50% up to 90% that of the parental pestivirus in the same cell type.

"Transfected cell" means a cell containing an exogenously introduced nucleic acid molecule, and includes cells that are transiently transfected with the exogenous nucleic acid.

"Transformed cell" or "stably transformed cell" means a cell containing an exogenously introduced nucleic acid molecule which is present in the cytoplasm or nucleus of the cell and may be stably integrated into the chromosomal DNA of the cell.

"Virus" means a virion, virus particle or a viral genome.

A chimeric viral RNA according to the invention is designed to comprise a 5′ NTR, an ORF, and a 3′ NTR, at least one of which is a chimeric region containing two operably linked nucleotide sequences that are from the same region of a pestivirus and an HCV.

Pestivirus-specific sequences useful in the invention can be taken from the appropriate genomic region of any cytopathic or noncytopathic type I or type II BVDV isolate, classical swine fever virus (CSFV) isolate, or border disease viral isolate. For a list of pestiviruses, see Thiel, H.-J., P. G. W. Plagemann, and V. Moennig. 1996. Pestiviruses, p. 1059-1073. In B. N. Fields, D. M. Knipe and P. M. Howley (ed.), Fields Virology. Raven Press, New York. HCV-specific sequences can be taken from any strain or isolate of HCV, including but not limited to HCV-1, HCV-1a, HCV-1b, HCV-1c, HCV-2a, HCV-2b, HCV-2c, HCV-3a. Preferably, the parental pestivirus is a cytopathic strain of BVDV and the parental HCV strain is HCV-1.

The pestivirus- and HCV-specific sequences are operably linked in the chimeric region, meaning the sequences are arranged such that the resulting chimeric structure is functional in the context of replication of the pestivirus. For example, in one preferred embodiment the chimeric viral RNA comprises a chimeric 5′ NTR which comprises a BVDV-specific 5′ terminal sequence of 5′-(G/A)UAU and an IRES derived from HCV, with the ORF and the 3′ NTR consisting of a sequence from the same regions of BVDV. The BVDV-specific sequences at the 5′ terminus and in the ORF and 3′ NTR are chosen such that they are functional in the context of BVDV, meaning the chimeric viral RNA expresses the replication machinery of BVDV and this replication machinery is capable of replicating the chimeric RNA. In addition, translation of the BVDV ORF in the chimeric viral RNA is dependent upon a functional HCV IRES. The presence of a functional HCV IRES in this chimera allows the chimera to be used to screen for compounds that target the HCV IRES and thereby inhibit translation of the BVDV ORF as well as replication of the chimeric virus. Such compounds would be expected to also inhibit translation of the ORF in a wild-type HCV and consequently inhibit HCV replication.

Compounds that could be screened for anti-HCV activity using this and other HCV-BVDV 5′ NTR chimeras include but are not limited to antisense RNAs, RNA decoys that bind proteins involved in recognition of the HCV-specific sequences, ribozymes, and small molecule inhibitors of critical RNA-protein interactions. The use of such substances for therapeutic applications are known in the art. See, e.g., Amarzguioui M, et al., "Hammerhead ribozyme design and application." *Cell Mol Life Sci.* 1998 Nov;54(11):1175-202; Welch PJ, et al., "Expression of ribozymes in gene transfer systems to modulate target RNA levels.", *Curr Opin Biotechnol.* 1998 Oct;9(5):486-96; Bramlage B, et al. "Designing ribozymes for the inhibition of gene expression."; *Trends Biotechnol.* 1998 Oct;16(10):434-8; Gewirtz AM, et al. "Nucleic acid therapeutics: state of the art and future prospects."; *Blood.* 1998 Aug 1;92(3):712-36; Altman S., "RNase P in research and therapy." *Biotechnology* (N Y). 1995

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It is contemplated that a number of replication-competent chimeric structures can be made that allow the function of various HCV sequence elements and proteins to be studied and targeted in drug screening assays. For example, the invention includes replication-competent HCV-pestivirus chimeras having a chimeric ORF. One such chimeric ORF is one comprising an HCV sequence encoding the structural proteins and a pestivirus sequence

encoding the nonstructural proteins. It is believed that upon introduction into a cell, such a HCV-BVDV ORF chimera will produce HCV-like virus particles that will be released from the cell and capable of infecting cells normally infected by wild-type HCV, i.e., cells expressing an HCV receptor such as human CD81. Such ORF chimeras would be useful to screen compounds for drugs that inhibit formation, release or entry of HCV particles. In addition, ORF chimeras that produce virus particles containing at least one HCV structural protein would be useful as vaccines against HCV. Other ORF chimeras contemplated by the invention include, for example, chimeras comprising a pestivirus sequence encoding structural proteins and an HCV sequence encoding one or more nonstructural proteins such as the NS3 protease, NS4A cofactor, NS5A phosphoprotein/interferon resistance determinant and/or the NS5B polymerase. Replication of such ORF chimeras would be dependent upon the function of the HCV nonstructural protein(s) and these ORF chimeras could be used to screen for drugs that target the HCV nonstructural protein(s) as well as to screen for and map potential drug resistance mutations in HCV nonstructural proteins. In addition, HCV-pestivirus ORF chimeras could be useful for developing alternative *in vivo* animal models for HCV replication and HCV-associated hepatocellular carcinoma to evaluate antivirals and anti-tumor agents.

The invention also provides replication-competent HCV-pestivirus chimeras having a chimeric 3′ NTR which contains one or more conserved elements of the HCV 3′ NTR. Such 3′ NTR chimeras would be useful for screening or evaluating compounds targeted against the HCV 3′ NTR. Compounds that could be screened include antisense RNA molecules, ribozymes and small molecule inhibitors of critical RNA-protein interactions. One 3′ NTR chimera according to the invention comprises a BVDV 5′ NTR, BVDV ORF and a chimeric 3′ NTR which consists of an HCV-specific sequence derived from the HCV 3′ NTR immediately followed by a BVDV 3′ NTR. The HCV-specific 3′ NTR that allows for replication in the context of BVDV has a deletion in the 3′ NTR poly (U) tract but has all the other HCV 3′ NTR elements, including the 98 bp 3′ terminal conserved element.

HCV-pestivirus chimeras included within the scope of the invention include those comprising combinations of chimeric regions, i.e., 5′ NTR and ORF chimeras; 5′ NTR and 3′ NTR chimeras; ORF and 3′ NTR chimeras; and chimeric RNAs in which each of the 5′ NTR, ORF and 3′ NTR regions comprise an HCV sequence operably linked to a pestivirus sequence.

The invention also provides chimeric RNAs having two ORFs, or bicistronic HCV-pestivirus chimeras. Bicistronic chimeras contemplated by the invention include structures in which the first ORF contains one or more HCV genes and is followed by a second IRES

operably linked to a second ORF encoding the pestivirus replicase machinery. It is also contemplated the first ORF may encode a heterologous sequence such as an antigen.

It is believed that many HCV-pestivirus chimeras of the invention will be attenuated as compared to the parental wild-type pestivirus. Such attenuated chimeric RNA genomes would be candidate vaccines in the form of live-attenuated virus particles or as RNA or cDNA "genetic" vaccines.

The invention also includes vaccines against HCV which comprise an immunogenically-effective amount of HCV-pestivirus particles or nucleic acid. Anti-HCV vaccines comprising virus particles should preferably contain one or more HCV structural proteins.

The therapeutic or pharmaceutical compositions of the present invention can be administered by any suitable route known in the art including for example by injection such as intraperitoneal, intravenous, subcutaneous, intramuscular, transdermal, intrathecal or intracerebral injection. Administration can be either rapid as by injection or over a period of time as by slow infusion or administration of slow release formulation.

Compositions according to the invention can be employed in the form of pharmaceutical or veterinary preparations. Such preparations are made in a manner well known in the pharmaceutical and veterinary arts. One preferred preparation utilizes a vehicle of physiological saline solution, but it is contemplated that other pharmaceutically acceptable carriers such as physiological concentrations of other non-toxic salts, five percent aqueous glucose solution, sterile water or the like may also be used. It may also be desirable that a suitable buffer be present in the composition. Such solutions can, if desired, be lyophilized and stored in a sterile ampoule ready for reconstitution by the addition of sterile water for ready injection. The primary solvent can be aqueous or alternatively non-aqueous.

The carrier can also contain other pharmaceutically-acceptable excipients for modifying or maintaining the pH, osmolarity, viscosity, clarity, color, sterility, stability, rate of dissolution, or odor of the formulation. Similarly, the carrier may contain still other pharmaceutically-acceptable excipients for modifying or maintaining release or absorption or penetration across the blood-brain barrier. Such excipients are those substances usually and customarily employed to formulate dosages for parenteral administration in either unit dosage or multi-dose form or for direct infusion into the cerebrospinal fluid by continuous or periodic infusion.

It is also contemplated that certain formulations containing a chimeric virus according to the invention are to be administered orally. Such formulations are preferably encapsulated and formulated with suitable carriers in solid dosage forms. Some examples of suitable

carriers, excipients, and diluents include lactose, dextrose, sucrose, sorbitol, mannitol, starches, gum acacia, calcium phosphate, alginates, calcium silicate, microcrystalline cellulose, polyvinylpyrrolidone, cellulose, gelatin, syrup, methyl cellulose, methyl- and propylhydroxybenzoates, talc, magnesium, stearate, water, mineral oil, and the like. The formulations can additionally include lubricating agents, wetting agents, emulsifying and suspending agents, preserving agents, sweetening agents or flavoring agents. The compositions may be formulated so as to provide rapid, sustained, or delayed release of the active ingredients after administration to the patient by employing procedures well known in the art. The formulations can also contain substances that diminish proteolytic degradation and promote absorption such as, for example, surface active agents.

The specific dose is calculated according to the approximate body weight or body surface area of the patient or the volume of body space to be occupied. The dose will also be calculated dependent upon the particular route of administration selected. Such calculations can be made without undue experimentation by one skilled in the art. Exact dosages are determined in conjunction with standard dose-response studies. It will be understood that the amount of the composition actually administered will be determined by a practitioner, in the light of the relevant circumstances including the condition or conditions to be treated, the choice of composition to be administered, the age, weight, and response of the individual patient, the severity of the patient's symptoms, and the chosen route of administration. Dose administration can be repeated depending upon the pharmacokinetic parameters of the dosage formulation and the route of administration used.

Replication-competent HCV-pestiviruses are generated by choosing the HCV function or sequence element desired to be studied. The HCV sequence can be obtained from a plasmid clone of a partial or full HCV genome using PCR to amplify a target region containing the desired sequence or by restriction enzyme digestion. The HCV fragment is then inserted into the desired location of a clone of the pestivirus genome using standard techniques. Desired portions of the pestivirus genome may be deleted before or after addition of the HCV fragment. The recombinant genome is then transfected into a cell that supports replication of the parental pestivirus genome and their ability to replicate using standard assays. For example, replication can be assessed by virus-induced cytopathic effect; plaque formation; detection of viral antigens and/or viral RNA accumulation; and by plaque assay measuring released infectious virus. The inventors herein have found that the BVDV RNA replication machinery works in many cell types, including bovine, hamster, mouse and human cells. It has also been reported that BVDV RNAs can amplify in other cell types including human hepatoma lines and hepatocytes (Behrens SE, et al., *J Virol.* 1998 Mar;72(3):2364-72).

The host cell range for a particular chimera will be dependent upon the properties of that chimera as empirically determined.

As described below, some chimeras do not replicate stably as indicated by heterogeneity in the size of plaques produced by the chimeric virus. Upon passage, pseudorevertants can frequently be isolated that are capable of stable replication. Such pseudorevertants will have one or more deletions or base substitutions in the HCV and/or pestivirus sequences. Information derived from these gain-of-function mutations can be used to define the elements necessary for generating stable, replication-competent chimeras of HCV and a pestivirus.

The invention provides a method for screening compounds for antiviral activity against HCV. The method involves comparing a test compound's effect on replication of a chimeric HCV-pestivirus RNA molecule as described above with the compound's effect on replication of the parental pestivirus. Compounds which have a greater effect on replication of the chimeric virus than the pestivirus are likely directed against the HCV portion of the chimera. Typically, the method is performed by providing duplicate cell cultures containing a chimeric viral RNA which is replication-competent in that cell, treating one of the culture with the test compound, and then measuring the replication efficiency of the chimeric RNA in both cultures. Any effect induced by the compound is compared against the compound's effect on replication of the parental pestivirus in cells of the same type. This control assay is preferably performed at the same time using the same culture conditions.

The cells used in the screening assay can be prepared by transiently transfecting the cells with the desired chimeric RNA molecule as described below. Alternatively, it is contemplated that the chimeric RNA molecule can be constitutively expressed in the cell by transfecting the cell with a polynucleotide comprising a cDNA of the chimeric RNA operably linked to a DNA-dependent promoter. The chimeric cDNA may include a selectable marker. which would allow for selection of cells expressing the chimeric RNA. It is also envisioned the selectable marker could be a dominant marker that allows selection of cells expressing chimeras having adaptive mutations or selection of cells permissive for virus replication (Frolov et al., *J. Virol.* 73:3854-3865, 1999). It is also contemplated the cDNA could express a reporter gene that could be assayed to measure RNA replication.

Alternatively, chimeric virus particles are incubated with a cell permissive for infection by the pestivirus in the presence or absence of the test compound and then replication of the chimeric virus is measured and compared to the replication of the parental pestivirus incubated with the same cell type in the presence or absence of the test compound.

Inhibition of replication can be measured in many ways, including assaying for the reduction of virus-induced cytopathic effect; inhibition of plaque formation, reduced production of viral antigens as detected by immunofluoresence assay; reduced viral RNA accumulation; reduction in released infectious virus from treated and untreated control and chimera samples using a plaque assay. In addition, it is contemplated that a cell line that is designed for pestivirus-specific transactivation of a reporter gene could be used directly or in lieu of a plaque assay. The reporter gene is operably linked to a promoter that is activated upon infection by the chimeric virus and production of the viral transactivator protein.

Preferred embodiments of the invention are described in the following examples. Other embodiments within the scope of the claims herein will be apparent to one skilled in the art from consideration of the specification or practice of the invention as disclosed herein. It is intended that the specification, together with the examples, be considered exemplary only, with the scope and spirit of the invention being indicated by the claims which follow the examples.

Example 1

This example illustrates the construction and analysis of 5′ HCV-BVDV chimeras as reported in detail in Frolov et al. (*RNA 4*:1418-1435, 1998) which is incorporated in its entirety by reference. A functional clone of BVDV (Mendez et al., *J. Virol. 72*:4737-4745, 1998) was used to construct and characterize a series of 5' NTR chimeras with sequences derived from HCV and the picornavirus, encephalomyocarditis virus (EMCV). The results help to define the requirements of a functional BVDV 5' NTR and provide replication-competent BVDV-HCV chimeras dependent on a functional HCV IRES.

Example 2

This example illustrates the construction of chimeras for expressing additional functional portions of the HCV genome by addition of further HCV sequence downstream from the functional or adapted HCV 5'NTR chimeras fused in-frame to the BVDV ORF.

One such construct (Figure 21) involves fusion of HCV sequences to BVDV sequences in the p7 protein coding region (at a convenient BseRI restriction site). Both HCV and BVDV encode a p7 protein that is located immediately downstream of the E2 protein. The p7 protein is a small hydrophobic protein of unknown function. pCBV/p7 consists of the first 79 bases of the BVDV 5'NTR encoding stem loop structure B1' and B1, followed by the entire HCV 5'NTR, the entire HCV structural protein coding region and the first 36 amino acids of HCV p7 fused to the C-terminal 31 amino acids of BVDV p7. The fused p7 gene is followed by the remainder of the BVDV ORF including the entire nonstructural region and the BVDV 3' NTR. Transfection of MDBK cells with the RNA corresponding to this

sequence (Figure 22) leads to replication of the chimeric RNA and production of the expected HCV and BVDV polyprotein cleavage products. Variations on this strategy are envisioned in which all or part of the HCV polyprotein and cis elements important for RNA packaging can be expressed in viable chimeras. In addition the BVDV replicase regions for either cytopathic or non-cytopathic pestiviruses (like NADL cIns-) can be used. Transfection of cells permissive for HCV particle, assembly, release and reinfection with this chimeric RNA can be used to make HCV-like particles. These particles and this infection system can be used (i) to screen for specific inhibitors of HCV particle, assembly, release and reinfection, (ii) for identifying antibodies capable of neutralizing HCV infectivity and (iii) as live or inactivated vaccines. Furthermore, this embodiment of the invention demonstrates that the BVDV RNA replication machinery can be used for expression of heterologous RNA and polypeptide sequences and can be used as a vehicle for RNA or DNA "genetic" vaccination in which the BVDV replicase amplifies the level of antigen expression by cytoplasmic RNA-dependent replication.

Example 3

This example illustrates chimeric RNA's that are modified to express dominant selectable markers, assayable markers or FACS sortable markers.

Such variants can be used to select for chimeras capable of replication in particular cell types, or to screen for cell types that are permissive for replication of the chimeric RNA. Selectable markers include, but are not limited to, the genes encoding puromycin resistance (puromycin N-acetyl transferase; PAC), neomycin resistance, blasticidin resistance, hygromycin resistance, etc. Assayable markers include, but are not limited to, the genes encoding B-galactosidase, luciferase, B-glucuronidase, etc. Easily sortable molecules include single chain antibodies, cell surface markers, and non-toxic protein markers like green fluorescent protein. In a specific example (Figures 23 and 24), the RNA encoded by pCBV/p7 was modified to include a cassette at the beginning of the BVDV 3'NTR that is comprised of the EMCV IRES driving the gene encoding PAC. This chimeric RNA can replicate, expresses PAC and confers resistance to puromycin resistance. This property can be used to select for variants of the chimera that are capable of noncytopathic replication in desired cells type and also provides a means of showing that cells harbor a functional chimeric RNA. Desired variants can be identified, cloned and further characterized as described in Example 1. Of note, is that this location in the BVDV genome and this strategy for expressing heterologous genes may also be applied to using infectious attenuated

pestiviruses as gene expression vectors and as chimeric live vaccines against other animal pathogens.

Example 4

This example illustrates the use of the bicistronic strategy as an alternative to the in-frame fusions described in Example 2.

A specific example is shown in Figure 25 and its sequence as Figure 26. In this bicistronic chimera, the 5' sequences are identical to that of pCBV/p7 except that the HCV ORF continues to include the first 246 amino acids of NS4B. The HCV sequence is followed by the EMCV IRES fused to BVDV Npro, the N-terminal 10 aa of BVDV C, the C-terminal 19 aa of C, 9 N-terminal amino acids of Erns, 48 C-terminal amino acids of E2 and the remainder of the BVDV NADL ORF and 3' NTR. The constructed BVDV ORF encodes a functional BVDV RNA replicase. The deletions in the N-terminal portion of this ORF were designed to preserve proper membrane topology and processing of the replicase. The bicistronic chimeric RNA can replicate upon transfection of permissive BVDV host cells.

Example 5

This example illustrates 3'NTR chimeras. Although initial attempts to recover viable chimeric viruses in which the BVDV 3'NTR was completely replaced by that of HCV were unsuccessful, a strategy similar to that detailed in Example 1 has produced chimeras that harbor the conserved elements of the HCV 3'NTR. An initial tandem 3'NTR construct was made in which the HCV 3'NTR was engineered to follow the BVDV ORF. The complete BVDV 3'NTR was position 3' to the HCV 3' NTR after a short heterologous sequence. This sequence of this parental construct, which replicated poorly, is shown in Figure 19 RNAs transcribed from this plasmid were of low specific infectivity suggesting that revertants or pseudorevertants might have arisen. Indeed isolation and sequence analysis of several independent plaque-forming variants revealed that deletions in the HCV poly U tract of various lengths had occurred. These revertant sequences are shown in Figure 20. When these altered HCV 3'NTRs were reconstituted into the original tandem 3' NTR parent, they gave rise to plaque forming RNA transcripts of high specific infectivity, demonstrating that these alterations restored the ability of the chimeric RNA to replicate. Large deletions in the U tract gave rise to virus with more robust replication and larger plaques while stably maintaining the conserved HCV 3'NTR 98-base element and the polypyrimidine "transition" region. Such

chimeric viruses can now be used to screen and evaluate antisense, ribozyme, and other therapeutics targeted against this conserved HCV RNA element that is essential for replication.

Materials and Methods

Plasmid Constructs

pACNR/BVDV NADL was previously described (Mendez et al., 1998, *supra*). pBVDV is a derivative of pACNR/BVDV NADL which contains a G→T transversion at nt 14994 that creates an *Xba* I site upstream of the T7 promoter (T. Myers & C.M. Rice, unpubl.). To facilitate construction of the chimeras, subclones were created. First, two fragments were isolated by PCR amplification of p90/HCVFLlongpU (Kolykhalov et al., *Science 277*:570-574, 1997) with primers #498 (5'-TGTACATGGCACGTGCCAGCCCC) and #498 (5'-GATCAACTCCATGGTGCACGGTCT) and pBVDV with primers #481 (5'-AGACCGTGCACCATGGAGTTGATC) and #482 (5'-CGTTTCACACATGGATCCCTCCTC). These two fragments were digested with *Apa*L I and ligated to produce a fragment containing a fusion of the HCV 5' NTR to the BVDV ORF. This fragment was digested with *Sac*I and ligated into pGEM3Zf(-) which had been digested with *Sma* I and *Sac* I to produce the subclone pGEM498-SacI. Next, a fragment containing the BVDV 5' NTR was synthesized by PCR amplification of pBVDV with primers #183 (5'-TTTTCTAGATAATACGACTCACTATAGTATACGAGAATTAGAAAAGGCACTCG) and #480 (5'-GGGGGCTGGCACGTGCCATGTACA). This fragment was digested with *Xba* I and *Bsr*G I and ligated into pGEM498-SacI digested with the same two enzymes, to create the plasmid pGEMXbaI-SacI. pGemXbaI-SacI contains a tandem fusion of the BVDV 5' NTR, the HCV 5' NTR, and the 5' portion of the BVDV N^{pro} gene. pBVDV + HCV was created by digesting pGEMXbaI-SacI with *Xba* I and *Sac* I and ligating the fragment into pBVDV digested with the same two enzymes, and as such pBVDV + HCV contains the T7 promoter, followed by the entire 385-nt 5' NTR of BVDV, a GT dinucleotide (nt 386-387), the entire 341-nt 5' NTR of HCV (nt 388-728), and the sequence of the BVDV NADL strain including the ORF and 3' NTR. Derivatives of pBVDV + HCV containing deletions within the BVDV 5' NTR and/or the HCV 5' NTR were created in the subclone pGEMXbaI-SacI, as described below, prior to ligation into *Sba* I- and *Sac* I-digested pBVDV. For making deletions, restrictions sites with non-compatible protruding ends were treated with the Klenow fragment of DNA polymerase I prior to ligation. For creation of pBVDV + HCVdelB3 (deletion of nt 174-374, inclusive), pGEMXbaI-SacI was digested with *Afl* II and *Bsr*G I. For pBVDV + HCVdelB2B3 (deletion of nt 67-374), pGEMXbaI-SacI was digested

with *Avr* II and *Bsr*G I. For pBVDV + HCVdelB1B2B3 (deletion of nt 33-374), pGEMXbal-Sacl was digested with *Sna*B I and *Bsr*G I. For pBVDV + HCVdelB2B3H1 (deletion of nt 67-3396), pGEMXbal-Sacl was digested with *Avr* II and *Xcm* I. For pBVDV + HCVdelB2B3H1H2 (deletion of nt 67-513), pGEMXbal-Sacl was digested with *AVR* II and *Bsg* I. For pBVDV + HCVdelB2B3H3 (deletion of nt 67-374, 518-704), subclone pGEMXbal-SacidelB2B3 was digested with *Sma* I. p5'HCV was created by digesting p90/HCVliongpU with *Xba* I and *Nru* I and ligating the fragment into pBVDV + HCV digested with the same two enzymes.

The EMCV plasmid, pEC_8, was provided by Ann Palmenberg and is described elsewhere (Hahn et al., *J. Virol 69*:2697-2699, 1995). p5'EMCV contains the entire 710 nt of the 5' NTR of EMCV, followed by the open reading frame of BVDV and the 3' NTR. One extra G residue was added between the T7 promoter and the first nucleotide of the EMCV 5' NTR to facilitate efficient in vitro transcription. Convenient restriction sites within the BVDV 5' NTR or the EMCV 5' NTR were used to create additional chimeras. Sites with noncompatible protruding ends were treated with the Klenow fragment of DNA polymerase I prior to ligation. For example, the plasmid pBVDV + EMCVdelA contains nt 1-378 of BVDV 5' NTR fused with nt 45-710 of EMCV (the *Bsr*G I site of BVDV ligated to the *Eco*R V site of EMCV), pBVDV + EMCVdelB3A contains nt 1-173 of BVDV fused with nt 45-710 of EMCV (the *Afl* II site of BVDV ligated to the *Eco*R V site of EMCV). pBVDV + EMCVdelB2B3A contains nt 1-66 of BVDV fused with nt 45-710 of EMCV (the *Avr* II site of BVDV ligated to the *Eco*R V site of EMCV). pBVDV + EMCVdelB3ABC contains nt 1-173 of BVDV fused with nt 161-710 of EMCV (the *Afl* II site of BVDV ligated to the *Psp*1405 site of EMCV). pBVDV + EMCVdelB2B3ABC nt 1-66 of BVDV fused with nt 161-710 of EMCV (the *Avr* II site of BVDV ligated to the *Psp*1406 site of EMCV). pBVDV + EMCVdelB3A-H contains nt 1-101 of BVDV fused with nt 289-710 of EMCV (the *Nhe* I site of BVDV ligated to the *Avr* II site of EMCV). pBVDV + EMCVdelB2B3A-H contains nt 1-62 of BVDV fused with nt 289-710 of EMCV (the *Avr* II site of BVDV ligated to the *Avr* II site of EMCV). The schematics of the chimeric 5' NTRs are presented in Figures 2 and 4.

All other heterologous 5' NTRs used in the study were generated by PCR using an oligonucleotide complementary to nt256-272 of the HCV 5' NTR and primers containing the sequence of the *Xba* I restriction site followed by the T7 promoter, the heterologous sequences found in sequenced pseudorevertants, or sequences corresponding to different regions of the HCV 5' NTR. All the fragments were subcloned into the plasmid, pRS2 (a derivative of pUC19), sequenced, and recloned into the p5'HCV plasmid by replacing the

fragment between the *XBa* I site located upstream of the T7 promoter and the *Nhe* I site (nt 249-254) in the 5' NTR of HCV.

Cell cultures

MDBK cells were obtained from M. Collett (ViroPharma, Inc.) and BT cells were obtained from the American Type Culture Collection (Rockville, Maryland). Cells were grown in Dulbecco's modified Eagle medium (D-MEM) supplemented with 10% horse serum and sodium pyruvate.

Transcriptions and transfections

All the designed plasmids , including pBVDV and the chimeric derivatives, were digested to completion with *Sda* I (*Sse*83871), purified by phenol extraction, precipitated by ethanol, and dissolved in water. The transcription reactions were performed sin the T7 Megascript kit (AMBION) using the conditions recommended by the manufacturer. Reactions were incubated at 37°C for 1 h, and ^{3}H-UTP was added to the reaction to quantify the RNA synthesis. The quality of the synthesized RNAs was checked by agarose gel electrophoresis, and samples containing 50-60% of full-length RNA were used for electroporations and in vitro translations. The reaction mixtures were aliquoted and stored at -70°C prior to electroporation or in vitro translations.

Transfection was performed by electroporation of MDBK cells using previously described conditions (Mendez et al., 1998, *supra*). Two micrograms of in vitro synthesized RNA, corresponding to approximately 1 μ g of the full-length transcript, were used per electroporation. In standard experiments, ten-fold dilutions of electroporated cells were seeded in 6-well tissue culture plates containing 5 x 10^5 naive MDBK cells per well. After 1 h of incubation at 37°C in an 5% CO_2 incubator, cells were overlaid with 3 ml of 0.6% LE Sea Kem agarose (FMC Bioproducts) containing minimal essential medium supplemented with 5% horse serum. Plaques were stained with crystal violet after 3 days incubation at 37°C. The rest of the transfected cells was seeded into 100-mm dishes and incubated for approximately 48 h or until cytopathic effect was observed in virtually all cells. Samples of the media were taken at 24 and 48 h, and virus titers were determined as described above and previously (Mendez et al., 1998, *supra*).

Analysis of the 5' ends of viral genomes

Sequencing of the 5' ends of selected variants of BVDV was performed on plaque-purified viruses. Plaques were typically isolated from the agarose overlay without staining with neutral red. Virus was eluted in 1 ml of D-MEM/10% horse serum for several hours and was used to infect 5 x 10^5 MDBK cells in 35-mm dishes. After 1 h of virus adsorption of 37

°C, an additional 1 ml of D-MEM/10% horse serum was added to the dishes, and incubation was continued for 36-48 h until cytopathic effect was observed in virtually all cells.

Fifty microliters of harvested viral stocks were clarified by low speed centrifugation, and viral RNAs were isolated by TRIzol reagent (Gibco-BRL) using the protocol recommended by the manufacturer. Sequencing of the 5' termini was performed using an oligonucleotide/cDNA-ligation strategy described elsewhere (Troutt et al., *Proc. Natl. Acad. Sci. USA 89*:9823-9825, 1992). The primer S1 (5'-GTCGTTTCACACATGGATCC), complementary to nt 710-729 of the BVDV genome, was used for cDNA synthesis. A phosphorylated oligonucleotide tag (5'-GACTGTTGTGGCCTGCAGGGCCGAATT) with an amino group on the 3' terminus was ligated to the first strand cDNA (Troutt et al., 1992, *supra*). One tenth of this reaction mixture was used for PCR amplification. The primers for PCR amplification were as follows: primer A (5'-GCCCTGCAGGCCACAACAGTC), complementary to the tag; primer B (5'-TCAGGCAGTACCACAA) complementary to nt 281-296 of the HCV 5' NTR; and primer C (5'-GGAATGCTCGTCAAGAAGACAG), complementary to nt 268-289 of the EMCV 5' NTR. The primer pairs of A + B or A + C were used for analysis of the pseudorevertants of 5'HCV and BVDV + HCVdelB1B2B3 or 5'EMCV, respectively. For the 5'HCV pseudorevertants, one tenth of the ligation mixture was used for an additional PCR reaction. This fragment was synthesized using primer S1, describe above, and a primer corresponding to nt 147-175 of the HCV genome. Fragments were purified by agarose gel electrophoresis and cloned into the plasmid pRS2. Multiple independent clones were sequenced by the standard dideoxy-mediated chain termination methods using the Sequenase version 2.0 DNA Sequencing Kit (USB).

Cell-free translation

Cell-free translation reactions were performed in reticulocyte extracts (Promega) using conditions recommended by the manufacture. Usually 0.1-1 μg of the same in vitro synthesized RNAs used in transfection experiments were used in 25 μl translation reactions. After 45 min of incubation at 30 °C, 2 μl were dissolved in 10 μl of sample buffer, and those samples were analyzed by sodium dodecyl sulfate PAGE. Labeled proteins were visualized by autoradiography of the dried gel. The efficiency of translation was measured using phosphorimager analysis (Molecular Dynamics) by comparing the radioactivity in the band corresponding to the N^{pro} protein. In preliminary experiments, an eightfold increase in incorporation was observed for translation of 4 μg versus 0.4 μg BVDV transcript RNA. Quantitative data were obtained from reactions using subsaturating (0.4 μg) amounts of BVDV or BVDV chimera transcript RNAs.

Analysis of virus specific RNAs

The protocols used for radioactive labeling of virus-specific RNAs are described in the appropriate figure legends. RNAs were isolated from the cells by using TRIzol reagent as recommended by the manufacturer (Gibco-BRL). After denaturation with glyoxal in dimethylsulfoxide, cellular RNAs were analyzed by electrophoresis in a 1% agarose gel containing a 10 mM phosphate buffer. Pieces of the dried gel containing the appropriate RNA bands were excised, and their radioactivity measured by liquid scintillation counting.

Results

Features of the BVDV, HCV, and EMCV 5' NTRs important for chimera design

Schematic representations of the proposed secondary structures of the 5' NTRs of HCV, BVDV, and EMCV are shown, and the location of each IRES is indicated in Figure 1. EMCV is a member of the cardiovirus genus within the family *Picornaviridae*. While not a member of the *Flaviviridae*, EMCV is similar to HCV and BVDV in that it is a positive-strand RNA virus shown to contain an IRES within its 5' NTR (Jang et al., *J. virol 62*:2636-2643, 1988). Based on their proposed secondary structures, the HCV IRES and the BVDV IRES have been classified as type 3 IRESs, while the EMCV IRES is classified as a type 2 IRES (Lemon & Honda, *Siemin. Virol. 8*:274-288, 1997). However, these three IRESs as well as IRESs from other members of the *Flaviviridae* and the *Picornaviridae* have been proposed to contain a common structural core (Le et al., *Virus Genes 12*:135-147, 1996).

The model for the secondary structure of the 341-nt HCV 5' NTR has been refined by enzymatic and chemical analysis of synthetic transcripts (Brown et al., *Nucl. Acids. Res. 20*:5041-5045, 1992; Wang et al., *J. Virol 68*:7301-7307, 1994; Honda et al., *RNA 2*:955-968, 1996; Lima et al., 1997). This element contains four discreet hairpins (referred to here as H1, H2, H3 and H4) and a pseudoknot at the base of hairpin H3 (Wang et al., 1995). The secondary structure of the 385-nt BVDV 5' NTR has not been as extensively studied, but is proposed to be similar to that of HCV (Brown et al., 1992) with four discrete hairpins (referred to here as B1', B1, B2, and B3) and a pseudoknot at the base of B3 (Rijnbrand et al., 1997). The secondary structure of the longer (>700 nt) EMCV 5' NTR consists of a series of hairpins A-M (Duke et al., 1992; Hoffman & Palmenberg, 1996). Recently, a revised model of the EMCV 5' NTR suggests moderately different secondary structures for the C and G subregions, and significantly different secondary structures for the I-M subregion (Palmenberg & Sgro, 1997).

For HCV, H1 is nonessential for IRES function (Reynolds et al., 1995; Rijnbrand et al., 1995; Honda et al., 1996b; Reynolds et al., 1996; Kamoshita et al., 1997) and its deletion

has actually increased translation efficiency in some analyses (Rijnbrand et al., 1995; Honda et al., 1996b). Most studies have found that hairpin H2 and H3 and the pseudoknot are essential for IRES function (Wang et al., 1993; Rijnbrand et al., 1995; Honda et al., 1996b). However, two studies indicate that H2 may not be essential (Tsukiyama-Kohara et al., 1992; Urabe et al., 1997). The 3' boundary of the HCV IRES is more controversial. The IRES clearly extends to the AUG initiation codon. However, some studies indicate that sequences affecting the efficiency of translation initiation extend into the ORF (Reynolds et al., 1995; Honda et al., 1996a; Honda et al., 1996b; Lu & Wimmer, 1996). By analogy to the HCV IRES and the related pestivirus CSFV IRES, the BVDV IRES probably requires hairpins B2 and B3 and the pseudoknot for function, with B1' and B1 probably not required for IRES activity (Poole et al., 1995; Rijnbrand et al., 1997). For EMCV, hairpins H-L have been shown to be required for IRES function in mono- or dicistronic constructs (Jang & Wimmer, 1990; Duke et al., 1992). The remaining portion of the EMCV 5' NTR is thought to be required for RNA replication or unknown steps in viral replication that are important for pathogenesis (Duke et al., 1990; Martin & Palmenberg, 1996).

Replacement of the BVDV 5' NTR with the HCV 5' NTR results in a large decrease in specific infectivity

Since the BVDV 5' NTR and the HCV 5' NTR are proposed to have similar RNA secondary structure and functional organization, an experiment was performed to test whether the BVDV 5' NTR could be replaced by the HCV 5' NTR. p5' HCV has an exact replacement of the BVDV 5' NTR with that of HCV (Fig. 2A) while the coding sequence and 3' NTR of p5'HCV are identical to pBVDV. Positioning of the HCV 5' NTR in such a manner was necessary since translation initiation from the HCV IRES begins at or near the AUG start codon (Honda et al., 1996a; Reynolds et al., 1995; Reynolds et al., 1996; Rijnbrand et al., 1996). The specific infectivity of 5'HCV RNA synthesized in vitro was compared to that of BVDV RNA by transfection of MDBK (bovine kidney) cells.(Fig. 2A). The specific infectivity of BVDV RNA was approximately 4 x 10^6 plaque forming units (PFU)/µg RNA. In contrast, the specific infectivity of 5' HCV RNA was near the limit of detection (30-50 PFU/µg RNA) and considerable plaque heterogeneity was apparent. These results suggested that the HCV 5' NTR replacement chimera might be incapable of efficient replication and plaque formation and that the plaque forming virus observed had arisen by secondary mutation(s). Sequence analysis of plaque-purified 5' HCV viruses presented below confirmed that the replicating pool of virus contained such pseudorevertants.

Next, the *in vitro* translation efficiency of these two RNAs in rabbit reticulocyte extracts was analyzed to test whether the defect in specific infectivity of 5' HCV RNA could be attributed to lower translation efficiency. Although the specific infectivity of 5' HCV RNA was reduced ~5 logs compared to BVDV RNA, its translation efficiency was only slightly reduced, ~twofold (Fig. 3, lane 1 vs. lane 2). The apparent size of the N-terminal cleavage product, N^{pro}, was identical for both RNAs, suggesting that translation initiated with the correct AUG. These data are consistent with the hypothesis that the BVDV 5' NTR contains signals that are required for a step in replication other than translation which are not present in the 5' HCV chimera.

Given the low specific infectivity of 5' HCV RNA, an experiment was performed to test the effect of placing the BVDV 5' NTR sequence upstream of the HCV 5' NTR, resulting in tandem BVDV and HCV 5' NTRs (called BVDV + HCV). This arrangement actually decreased translation efficiency (Fig. 3, lane 14 vs. lane 1) yet restored infectivity (Fig. 2A). The plaques produced by BVDV + HCV were also heterogeneous in size, indicating that this virus was unstable. Upon passage, RT-PCR analysis indicated that pseudorevertants had indeed arisen in which portions of the BVDV and/or HCV 5' NTRs had been deleted (data not shown). These data show that sequences in the BVDV 5' NTR required for virus replication can function when placed upstream of a functional HCV IRES driving translation of the BVDV polyprotein.

Hairpins B1' and B1 in conjunction with the HCV IRES are sufficient for stable and efficient BVDV replication

The sequences within the BVDV 5' NTR that restored replication in the context of the HCV 5' NTR were mapped using three deletion variants. The deletion BVDV + HCVdelB3 removed a large portion of hairpin B3; the deletion within BVDV + HCVdelB2B3 removed hairpins B2 and B3, and the deletion within BVDV + HCVdelB1B2B3 removed hairpins B1, B2 and B3. The specific infectivities of RNAs from these deletion mutants were near that of BVDV RNA (Fig. 2). Upon passage of these viruses, RT-PCR analyses and sequencing indicated that BVDV + HCV delB3 and BVDV + HCVdelB2B3 were stably propagated and produced homogeneous plaques slightly smaller than those of wild-type BVDV (data not shown). In contrast, BVDV + HCVdelB1B2B3 produced smaller heterogeneous plaques. Reverse transcription-polymerase chain reaction (RT-PCR) analysis and sequencing indicated that BVDV + HCVdelB1B2B3 underwent a reversion event described in more detail below. The translation efficiencies of these three RNAs (Fig. 3, lanes 9, 10, and 12) were similar to BVDV + HCV RNA (Fig. 3, lane 14), indicating that the deleted portions (hairpins B1, B2,

and B3) are not required for translation in the BVDV + HCV chimera. These results show that B1' and B1 are the minimal elements sufficient for stable replication in conjunction with the HCV 5' NTR.

Having shown that B1' and B1 are sufficient for replication in conjunction with the HCV 5' NTR, we next conducted a deletion analysis to determine the sequences within the HCV 5' NTR of BVDV + HCV delB2B3 required for replication. A large portion of H1 was deleted in BVDV + HCV delB2B3H1, while both H1 and H2 were deleted in BVDV + HCV delB2B3H1H2. Of these two RNAs, only BVDV + HCV delB2B3H1 was as infectious as parental BVDV RNA (Fig. 2B). However, the BVDV + HCV delB2B3H1 virus produced smaller plaques than BVDV + HCV delB2B3, indicating that hairpin H1 may augment replication of the chimera. In contrast, BVDV + HCV delB2B3H1H2 RNA was not infectious (Fig. 2B) and was translated poorly (Fig. 3, lane 11). Diminished HCV IRES activity might be due to deletion of hairpin H2 or juxtaposition of BVDV hairpins B1' and B1 with H3. A third derivative of BVDV + HCV delB2B3, with a *Sma* I-*Sma* I deletion abrogating HCV IRES function by removing H3, was also not infectious (data not shown). Thus, a 5' NTR consisting of B1' and B1 and a functional HCV IRES is sufficient for stable BVDV replication in MDBK cells. Similar results were obtained in BT cells, another BVDV-permissive continuous bovine cell line (data not shown).

Replacement of the BVDV 5' NTR with the EMCV 5' NTR

The following experiment was performed to determine whether the BVDV 5' NTR could be replaced by the 5' NTR of a more phylogenetically distant virus, EMCV. A derivative of BVDV was created, called 5' EMCV, that contains an exact replacement of the BVDV 5' NTR with the EMCV 5' NTR plus an additional guanosine residue at the 5' terminus for more efficient transcription initiation of T7 polymerase (Fig. 4A). The specific infectivity of 5' EMCV RNA was more than three orders of magnitude lower than BVDV RNA, indicating that it was defective for replication, although its specific infectivity was higher than that of 5' HCV RNA (compare Figs. 4A and 2A). Similar to 5' HCV, 5' EMCV produced heterogeneous plaques, and sequence analysis indicated that pseudorevertants had arisen. The lower specific infectivity of 5' EMCV RNA was not likely because of a defect in translation, since the translation efficiency of 5' EMCV RNA was about threefold higher in vitro than that of BVDV RNA (Fig. 3, lane 20 vs. lane 19).

Similar to BVDV + HCV, it was also determined whether the BVDV 5' NTR at the 5' end of the 5' EMCV RNA would increase its specific infectivity. BVDV + EMCVdelA (Fig. 4A) contained the entire BVDV 5' NTR in tandem with the EMCV 5' NTR lacking a portion

of hairpin A. BVDV + EMCVdelA RNA had a specific infectivity near that of BDVD RNA (compare Figs. 4A and 2A) despite having a lower translation efficiency than 5' EMCV (Fig. 3, lane 21 vs. lane 20). Similar to the results with BVDV + HCV, this implicates the added BVDV 5' NTR sequence for a step in viral replication other than translation. Two derivatives of BVDV + EMCVdelA that contain deletions of portions of the BDVD 5' NTR but maintain the sequence of B1' and B1, BDVD + EMCVdelB3A and BVDV + EMCVdelB2B3A (Fig. 4A), also were infectious. These derivatives had translation efficiencies near that of the parental BVDV + EMCVdelA (Fig. 3, compare lanes 15 and 16 with lane 21). This demonstrated that hairpins B1' and B1 were sufficient for replication in conjunction with a large portion of the EMCV 5' NTR. Derivatives of BVDV + EMCVdelB3A or BVDV + EMCVdelB2B3A that contain further deletions of EMCV (BVDV _ EMCVdelB3ABC and BVDV + EMCVdelB2B3ABC in particular) were translated efficiently (Fig. 3, lanes 17 and 18) and were infectious (Fig. 4B). This indicates that the chimeras did not require putative EMCV RNA replication signals (Martin & Palmenberg, 1996). However, derivatives with deletions extending into the canonical EMCV IRES were not infectious. For example, BVDV + EMCVdelB3A-H and BVDV + EMCVdelB2B3A-H, in which a portion of hairpin H is deleted, were not infectious (Fig. 4B) and were inefficiently translated in vitro (Fig. 3, lanes 22 and 23). It should be noted that all of the BVDV + EMCV chimeras produced plaques of heterogeneous size, indicating some instability.

Relatively simple 5' NTR mutations are observed in adapted pseudorevertants

As mentioned previously, BVDV + HCVdelB1B2B3 did not replicate stably as indicated by the heterogeneity in the size of plaques produced by this virus. Upon passage and selection of medium plaque-producing variants, 5' RACE analysis and sequencing indicated that nt 1-26 had been deleted in the pseudorevertants, removing a large portion of B1' which was apparently deleterious in the absence of B1. This deletion results in the 5' terminal sequence 5'GUAUCG which is identical to the first six bases of BVDV genome RNA (Fig. 5) and is repeated at positions 27-32.

Analysis of the passaged 5' EMCV virus indicated that the replicating progeny had also undergone a simple deletion of sequence at the 5' end to generate more efficiently replicating variants (Fig. 5). After electroporation, the 5' EMCV virus pool was passaged 5 times at a multiplicity of infection of 0.1-1 PFU/cell on MDBK or BT cells, and the 5' termini of three randomly picked plaques were sequenced. For all three plaques selected, nt 2-209 had been deleted, again creating a genome RNA with the 5' terminal tetranucleotide sequence 5'-GUAU.

Analysis of the 5' HCV progeny indicated that more complicated variants had arisen. Most small plaque-producing variants were unstable and quickly reverted to medium plaque-producing variants. However, one small plaque-producing variant and two stable medium plaque-producing variants were isolated. 5' terminal sequences of the variants were amplified by rapid amplification of cDNA ends (RACE) and cloned into a plasmid vector, and sequences for several independent colonies were determined. The sequence of three clones of the small plaque-producing virus (5'HCV.R1) contained a deletion of HCV sequence from nt 1-34 and an addition of the dinucleotides 5'-AU in two clones and 5'-GU in the third clone. This creates a 5' terminus of 5'-(G/A) UAA (Fig. 5B), reminiscent of the first three bases of the BVDV genome RNA (5'-GUA). Both medium plaque variants appeared to have arisen by RNA recombination with non-viral sequences (Fig. 5). One medium plaque variant (5' HCV.R2) had deleted the first 21 bases of the HCV sequence and contained instead a heterologous sequence of 22 bases. BLAST searches revealed a perfect match between this sequence and a sequence in a human retina cDNA of unknown function (Tsp509I). The second medium plaque variant (5' HCV.R3) had also undergone a possible recombination event leading to the addition of 12 nt to the 5' end of the HCV sequence. Given its short length, multiple matches were found in the database with this sequence. As for the small plaque variant, sequencing of multiple clones revealed heterogeneity oat the extreme 5' end, with either G of A identified as the 5' base. Remarkably, for both medium plaque variants, the fused heterologous sequence began with the tetranucelotide sequence 5'-(G/A) UAU (Fig. 5B). For all three variants, sequencing of the entire 5' NTR and a portion of the N^{pro} coding region revealed only these changes at the 5' termini.

5' NTR sequence changes are sufficient for the pseudorevertant phenotypes

To assess the importance of these alterations oat the 5' terminus of the 5' HCV pseudorevertants, derivatives of 5' HCV were created with the changes determined by 5' RACE (Fig. 6A) and analyzed the specific infectivities of these RNAs (Fig. 6B). Corresponding to the small plaque variant, a derivative called 5' HCV.R1 orig was engineered which contained a 5' NTR consisting of the dinucleotide 5' -GU at the 5' terminus of HCV nt 35-341. This results in a 5' terminus consisting of 5'-GUAA. 5'HCV.R1 orig RNA had a specific infectivity at least four orders of magnitude higher than 5' HCV RNA (Figs. 6B and 2A). This demonstrates that this 5' NTR structure is sufficient for phenotypic reversion to high specific infectivity. However, small plaques and considerable heterogeneity were observed for 5'HCV.R1 orig suggesting that additional mutations may be present in the original small plaque variant.

The engineered derivative 5'HCV.R2orig had a 5' NTR consisting of 22 nt of Tsp509I-homologous sequence followed by HCV nt 22-341. Another construct, called 5'HCV.R3orig was made, which has the 12 nt of the other heterologous sequence fused to the intact HCV 5' NTR. Specific infectivities for both these derivatives were essentially the same as observed for wild type BVDV RNA (2-4 x 10^6 PFU/μg; Fig. 6B). Transfection with these transcripts produced medium plaques, as observed for the original variants, and this phenotype was stable upon passaging. These results show that the altered 5'NTR sequences were responsible for the pseudorevertant phenotypes rather than changes elsewhere in their genomes.

Addition of the tetranucleotide sequence 5'-GUAU to the HCV 5' NTR allows efficient BVDV replication

For all three 5' HCV variants studied, as well as the BVDV + HCV delB1B2B3 and 5'EMCV pseudorevertants, 5' NTR alterations seemed to involve creation of a three- or four-base "consensus" sequence identical to the 5' terminus of BVDV genome RNA. To test the importance of this sequence, as opposed to fused heterologous sequences, we created a set of variants with the BVDV 5' tetranucleotide sequence linked to the HCV 5' NTR or the deletion/recombinant break points identified during sequence analysis of the 5' HCV pseudorevertants (Fig. 6A). 5' HCV.R1cons had the tetranucleotide sequence 5'-GUAU fused to HCV nt 35-341. 5'HCV.R2cons had the 5'-GUAU tetranucleotide sequence fused to HCV nt 22-341. 5'HCV.R3cons contained the tetranucleotide sequence 5'-Guau fused to the intact 5' terminus of the HCV NTR. RNAs from all three of these derivatives had specific infectivities more than five orders of magnitude higher than 5'HCV and comparable to parental BVDV (Fig. 6B).

There were, however, significant differences between the phenotypes of some of these derivatives versus the reconstructed pseudorevertants. As mentioned above, 5'HCV.R1orig yielded tiny and small plaques and produced low virus yields even after 48 h. In contrast, the addition of four bases rather than two bases (5'-GUAU vs. 5'-GU) yielded virus with near wild-type plaque morphology (Fig. 6B) and growth Rates (Fig. 7). In the case of the smaller deletion, 5'HCV.R2orig and 5'HCV.R2cons were indistinguishable, suggesting that, other than the 5' four bases, the fused heterologous sequences were dispensable. This was not he case, however, for the chimera containing the 5'-GUAU tetranucleotide sequence

fused to the intact HCV 5' NTR. 5'HCV.R3cons produced small plaques (Fig. 6B) and grew more slowly than 5'HCV.R3orig (Fig. 7) suggesting that the sequence/structure of the sequences downstream of the 5' four bases can affect replication efficiency.

The tetranucleotide sequence 5'-GUAU is important for efficient BVDV RNA accumulation

Next, the effects of the different 5' termini on virus-specific RNA accumulation directly after transfection were analyzed. This allowed a direct comparison between 5'HCV and the reconstructed pseudorevertants as well as selected BVDV + HCV deletion constructs. MDBK cells were transfected with in vitro synthesized RNAs and labeled for 10 h beginning at 5 h post-transfection with ^{3}H-UTP in the presence of actinomycin D (Fig. 8). RNA replication of the 5' HCV chimera was severely impaired to a level below detection (Fig. 8, lane 2). In contrast, every 5' NTR alteration of 5' HCV that increased RNA specific infectivity and allowed efficient virus growth led to readily detectable viral RNA accumulation. Addition of B1' and B1 to the 5' terminus of the HCV 5' NTR restored RNA replication to a level ~50% of that observed for BVDV (BVDV + HCVdelB2B3; Fig. 8, lane 3 vs. lane 1). BVDV + HCVdelB2B3H1 displayed reduced RNA synthesis compared to BVDV + HCVdelB2B3 (Fig. 8, lane 4 vs. lane 3) perhaps explaining its small plaque phenotype and suggesting a possible positive role for H1 in replication of this chimera. 5'HCV.R1orig, which had exhibited plaque heterogeneity and slow growth, accumulated less RNA when compared to 5'HCV.R1cons (Fig. 8, lane 5 vs. lane 6). 5'HCV.R2orig and 5'HCV.R2cons showed similar RNA accumulation (Fig. 8, lane 9 vs. lane 10) consistent with their medium plaque phenotypes; and 5'HCV.R3cons exhibited reduced RNA synthesis compared to 5'HCV.R3orig (Fig. 8, lane 8 vs. lane 7), consistent with their small-versus medium-plaque phenotypes.

Although these RNA phenotypes are complex, the most striking result is that addition of the B1' B1 hairpins, addition of heterologous 5' sequences terminating with 5'-GUAU or simply fusion of this tetranucleotide sequence with the HCV 5' NTR or short 5' truncations of the HCV 5' NTR all dramatically upregulated RNA accumulation. This occurred without increasing translation efficiency, at least as measured in a cell-free assay (Fig. 3, compare lanes 3-8 to lane 1), suggesting that these sequences function at the level of RNA replication or stability.

Discussion

The work presented here helps to define the requirements for a functional BVDV 5'NTR. The BVDV-specific 5' NTR sequences required for efficient replication in cell culture are minimal and consist of the 5' terminal sequence, 5'-GUAU. The sequence 5'-AUAU, detected for some pseudorevertants, may also be functional but this was not tested for technical reasons. This simple 5'-terminal tetranucleotide sequence, which is conserved among pestivirses (Ruggli et al., 1996; Becher et al., 1998), was shown to function in the context of functional IRES elements derived from the hepacivirus HCV or the picornavirus EMCV. As discussed below, this may indicate that the 5' signals required for BVDV RNA replication are rather simple or that elements in these heterologous IRESs can functionally replace deleted BVDV sequences.

Sequences at the extreme 5' end of BVDV genome RNA could modulate the efficiency of RNA accumulation by affecting RNA stability, translation, promoter efficiency, or some combination of these processes. At this time, we can not distinguish among these possibilities but favor an effect on RNA replication. The complement of the BVDV 5' sequence at the 3' end of the negative-strand RNA presumably functions in the initiation of positive-strand RNA synthesis. Thus, AUAC-3' at the 3'terminus fo minus-strand RNA may be important for positive-strand RNA synthesis. Interestingly, for some positive-strand RNA viruses such as rubella virus (Pugachev & Frey, 1998), flock house virus (Ball, 1994) and turnip crinkle virus (Guan et al., 1997), only minimal *cis*-acting sequences at the 3' termini of negative-strand RNAs are required positive-strand RNA synthesis. In contrast to the 5' NTR replacements, we were unable to generate replication-competent BVDV-HCV replacing that of BVDV (data not shown). This may indicate that the signals within the pestivirus 3' NTR required for initiation of negative-strand RNA synthesis are more complex and virus specific. Once the replication complex has assembled at the 3' NTR and transversed the RNA during negative-strand synthesis, the requirements of the 5' NTR for initiation of positive-strand synthesis may be minimal.

Although the RNA replication signals within the 5' NTR appear to be rather simple, it is possible that the signals important for RNA replication actually extend into the IRES and are more complicated. For instance, the 5'HCV pseudorevertants were more stable and grew to higher titers than the 5'EMCV counterparts, despite the fact that the 5'EMCV RNAs were translated more efficiently in vitro. This may indicate that the BVDV and HCV IRESs contain signals important for RNA synthesis that are absent in the EMCV IRES.

It is perhaps not surprising that 5' HCV appeared to recombine with cellular mRNAs to acquire a 5' terminus with the 5' -(G/A) UAU consensus, given that non-cytopathic strains

of BVDV can recombine with BVDV RNA or cellular mRNAs to generate cytopathic strains of BVDV (Meyers & Thiel, 1996). Presumably, this recombination event involves template switching during negative-strand RNA synthesis, as observed for polio-virus (Kirkegaard & Baltimore, 1986). In contrast to 5' HCV, simple deletions of 5' terminal viral sequences could account for the BVDV + HCVdelB1B2B3 and 5'EMCV pseudorevertants since the tetranucleotide sequence is present in these 5' NTRs upstream of functional IRES elements. Such deletions could occur by partial degradation of positive-strand template prior to negative-strand synthesis, by premature termination during negative-strand RNA synthesis, or by degradation of 3' terminal negative-strand sequence after synthesis. It is proposed that 5'HCV was forced to recombine with cellular sequences because HCV does not have an 5'-(G/A) UAU sequence upstream of its IRES. The first occurrence of an (G/A)UAUA tetranucleotide sequence is at nt 94-97 within hairpin H2, and a 5' deletion extending into this sequence would presumably inactivate or severely impair HCV IRES activity. It is interesting that BVDV + HCVdelB1B2B3 and 5'EMCV pseudorevertants were generated at much higher frequency than 5'HCV pseudorevertants. This may indicate that recombination between BVDV and cellular RNAs is a rare event compared to the processes which lead to deletion of terminal viral sequences.

Poliovirus chimeras dependent upon a functional HCV IRES have been reported (Lu & Wimmer, 1996). Interestingly, viable poliovirus chimeras were produced only when HCV sequences included both the IRES and the N-terminal portion of the HCV ORF. Nucleotide sequences or structures in the downstream ORF can modulate HCV IRES translational efficiency (see Reynolds et al., 1995; Honda et al., 1996a) but it was also suggested that the N-terminal portion of the HCV core polypeptide might be involved. In the case of our 5' HCV pseudorevertants, there is no requirement for HCV C protein sequences. Although the translation efficiency of the HCV IRES in the presence of additional HCV sequences 3' to the AUG start was not directly assessed, the HCV chimeras and pseudorevertants were translationally active and infectious in the absence of any portion of the HCV ORF. This indicates that either the HCV IRES does not extend into the HCV ORF or that the BVDV ORF contains analogous sequence which functions in our 5'HCV chimeras. There is some limited identity between HCV and BVDV within this region. For example, HCV nt 359-394 and BVDV nt 405-440 are identical at 21 of 36 positions, although identity within this sequence may be attributed to a high adenosine content. It is interesting to note that the luciferase (LUC) and chloramphenicol acetyl transferase (CAT) reporter genes previously used to detect HCV IRES activity (Tsukiyama-Kohara et al., 1992; Wang et al., 1993) also have adenosine- or purine-rich regions in relatively the same position as the HCV ORF and

BVDV ORF. It this region is indeed important for IRES activity, this may explain why some have observed that the HCV IRES does not require a portion of the HCV ORF for translation of CAT or LUC (Tsukiyama-Kohara et al., 1992; Wang et al., 1993). Point mutations and insertions within this region of HCV have been shown to reduce HCV IRES activity in vitro (Honda et al., 1996a,b).

Despite the fact that B1' and B1 are conserved among different strains of BVDV and similar hairpins are present in border disease virus and CSFV (Deng & Brock, 1993; Becher et al., 1998), B1' and B1 were dispensable for BVDV replication, provided that the 5' tetranucleotide sequence 5'-(G/A)UAU remained. This may indicate a role for B1' and B1 in viral replication in vivo that we do not observe in cell culture. It will be interesting to test the phenotype of chimeras that lack B1' and B1 in vivo to determine if they are attenuated and might serve as useful BVDV vaccines. In this vein, several studies with flaviviruses have demonstrated that alterations in 5' NTR or 3' NTR elements can lead to attenuation in vivo (Cahour et al., 1995; Men et a., 1996; Mandl et al., 1998). BVDV chimeras that utilize the HCV or EMCV IRES may also prove to be attenuated simply due to the presence of the heterologous IRES. For poliovirus, it has been shown that differences in IRES efficiency in different host-cell environments can modulate host range and virulence (Shiroki et al., 1997).

BVDV-HCV chimeras that are dependent on a functional HCV IRES may have another practical application. It may be possible to use these chimeras to screen for anti-HCV therapeutics that target the HCV IRES. Other researchers have shown antisense oligonucleotide-mediated inhibition of HCV gene expression in hepatocytes by targeting the oligonucleotides to the HCV IRES (Hanecak et al., 1996). It will be of interest to measure the efficacy of antisense oligonucleotides or ribozymes (Lieber et al., 1996) against replicating virus, and these chimeras are more useful than HCV for this purpose since they are able to replicate efficiently in cell culture. BVDV is believed to be a reasonable model of HCV replication not only because of homology and conserved motifs within the 5' NTR but also because of similarities in overall genetic organization (Rice, 1996) and polyprotein processing strategy (Tautz et al., 1997; Xu et al., 1997).

In view of the above, it will be seen that the several advantages of the invention are achieved and other advantageous results attained.

As various changes could be made in the above methods and compositions without departing from the scope of the invention, it is intended that all matter contained in the above description and shown in the accompanying drawings shall be interpreted as illustrative and not in a limiting sense.

All references cited in this specification, including patents and patent applications, are hereby incorporated by reference. The discussion of references herein is intended merely to summarize the assertions made by their authors and no admission is made that any reference constitutes prior art. Applicants reserve the right to challenge the accuracy and pertinency of the cited references.

What is Claimed is:

1. A polynucleotide comprising a chimeric viral RNA which comprises:

(a) a 5′ nontranslated region (5′ NTR);

(b) an open reading frame (ORF) region; and

(c) a 3′ nontranslated region (3′ NTR);

wherein at least one of said regions is chimeric and comprises a first nucleotide sequence from a pestivirus in operable linkage with a first nucleotide sequence from an hepatitis C virus (HCV), and wherein said chimeric viral RNA is replication-competent.

2. The polynucleotide of claim 1, wherein the chimeric region is the 5′ NTR and the first pestivirus nucleotide sequence is from a bovine viral diarrhea virus (BVDV).

3. The polynucleotide of claim 2, wherein the BVDV nucleotide sequence is located at the 5′ terminus of the chimeric 5′ NTR and comprises 5′ RUAU.

4. The polynucleotide of claim 3, wherein the first HCV nucleotide sequence in the chimeric 5′ NTR comprises an internal ribosome entry site (IRES).

5. The polynucleotide of claim 4, wherein the ORF and the 3′ NTR consist of second and third BVDV sequences.

6. The polynucleotide of claim 5, wherein the 5′ terminal sequence comprises 5′ GUAU.

7. The polynucleotide of claim 4, wherein the ORF comprises a second HCV sequence encoding at least one structural protein operably linked to a second BVDV sequence.

8. The polynucleotide of claim 1, wherein the pestivirus is BVDV and the chimeric region is the 3′ NTR.

9. The polynucleotide of claim 8, wherein the first HCV sequence in the chimeric 3′ NTR comprises the HCV 98 bp 3′ terminal element (SEQ ID NO:X) operably linked to the first BVDV sequence.

10. A method for identifying compounds having antiviral activity against hepatitis C virus (HCV) comprising the steps of:

(a) providing a first cell containing a chimeric viral RNA which is replication-competent in the cell, the chimeric viral nucleic acid comprising a 5′ nontranslated region (5′ NTR), an open reading frame (ORF) region; and a 3′ nontranslated region (3′ NTR); wherein at least one of said regions is chimeric and comprises a first nucleotide sequence from a pestivirus in operable linkage with a first nucleotide sequence from an hepatitis C virus (HCV);

(b) providing a second cell containing the pestivirus; and

(c) comparing the replication efficiency of the chimeric viral RNA acid in the presence and absence of a test compound to the replication efficiency of the pestivirus in the presence and absence of the test compound,

wherein a greater reduction in compound-induced replication efficiency of the chimeric viral RNA than the pestivirus indicates the compound has anti-HCV activity.

11. The method of claim 10, wherein the chimeric region is the 5′ NTR and the first pestivirus nucleotide sequence is from a bovine viral diarrhea virus (BVDV).

12. The method of claim 11, wherein the BVDV nucleotide sequence is located at the 5′ terminus of the chimeric 5′ NTR and comprises 5′ RUAU.

13. The method of claim 12, wherein the first HCV nucleotide sequence in the chimeric 5′ NTR comprises an internal ribosome entry site (IRES).

14. The method of claim 13, wherein the ORF and the 3′ NTR comprise second and third sequences from the BVDV.

15. The method of claim 10, wherein the pestivirus is BVDV and the chimeric region is the 3′ NTR.

16. A genetically-engineered virus comprising a chimeric RNA genome which comprises:

(a) a 5′ nontranslated region (5′ NTR);

(b) an open reading frame (ORF) region; and

(c) a 3′ nontranslated region (3′ NTR);

wherein at least one of said regions is chimeric and comprises a first nucleotide sequence from a pestivirus in operable linkage with a first nucleotide sequence from an hepatitis C virus (HCV), and wherein said chimeric RNA genome is replication-competent.

17. The genetically-engineered virus of claim 16, wherein the chimeric region is the 5′ NTR and the first pestivirus nucleotide sequence is from a bovine viral diarrhea virus (BVDV).

18. The genetically-engineered virus of claim 16, wherein the BVDV nucleotide sequence is located at the 5′ terminus of the chimeric 5′ NTR and comprises 5′ RUAU and the first HCV nucleotide sequence in the chimeric 5′ NTR comprises an internal ribosome entry site (IRES).

19. A vaccine against bovine viral diarrhea virus (BVDV) comprising an immunogenically-effective amount of a genetically-engineered virus comprising a chimeric RNA genome having:

(a) a 5′ nontranslated region (5′ NTR);

(b) an open reading frame (ORF) region; and

(c) a 3′ nontranslated region (3′ NTR);

wherein at least one of said regions is chimeric and comprises a first nucleotide sequence from BVDV in operable linkage with a first nucleotide sequence from an hepatitis C virus (HCV), and wherein the genetically-engineered virus is attenuated as compared to BVDV.

20. The vaccine of claim 19, wherein the chimeric region is the 5′ NTR and the BVDV nucleotide sequence is located at the 5′ terminus of the chimeric 5′ NTR and comprises 5′ RUAU and the first HCV nucleotide sequence in the chimeric 5′ NTR comprises an internal ribosome entry site (IRES).

21. A polynucleotide comprising a chimeric viral RNA which comprises:

(a) a 5′ nontranslated region (5′ NTR);

(b) an open reading frame (ORF) region; and

(c) a 3′ nontranslated region (3′ NTR);

wherein at least one of said regions is chimeric and comprises a first nucleotide sequence from a pestivirus in operable linkage with a heterologous nucleotide sequence and wherein said chimeric viral RNA is replication-competent.

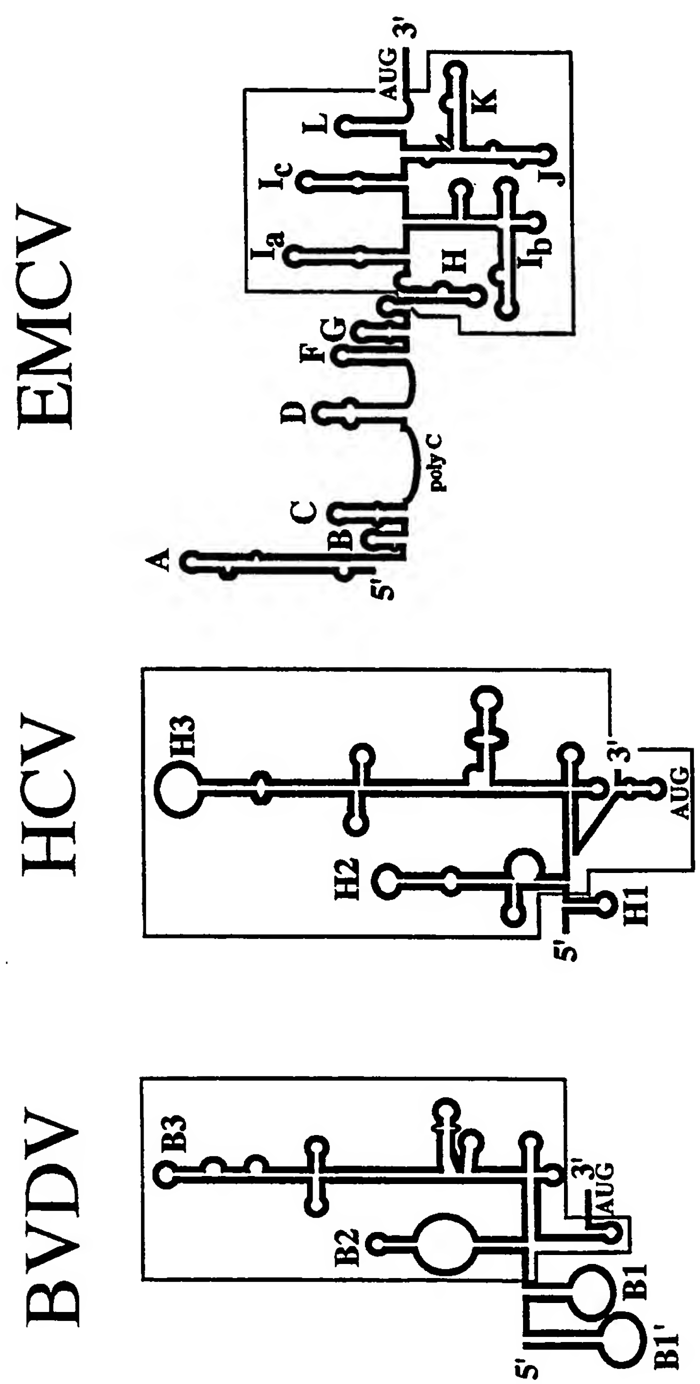
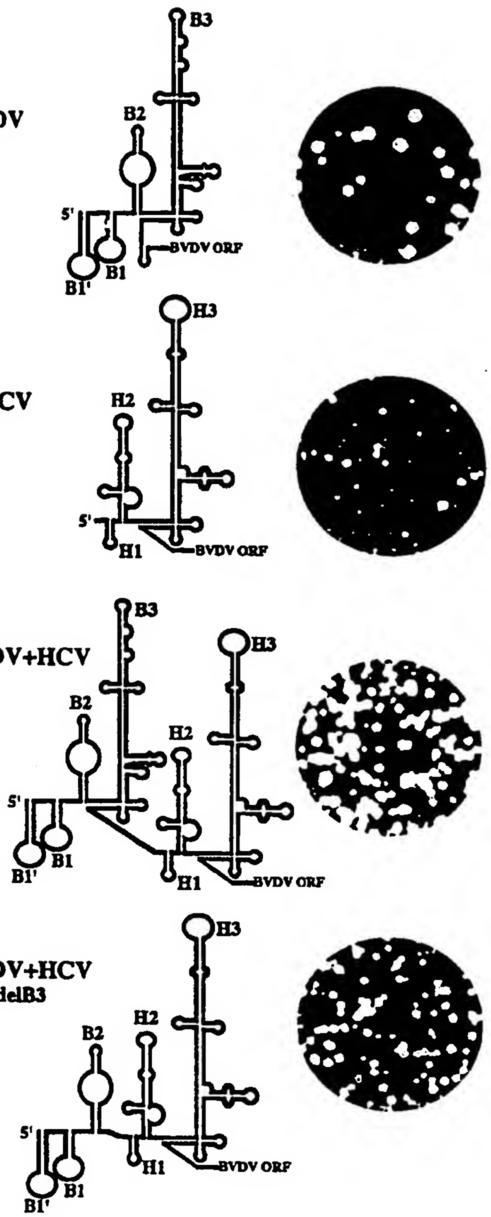
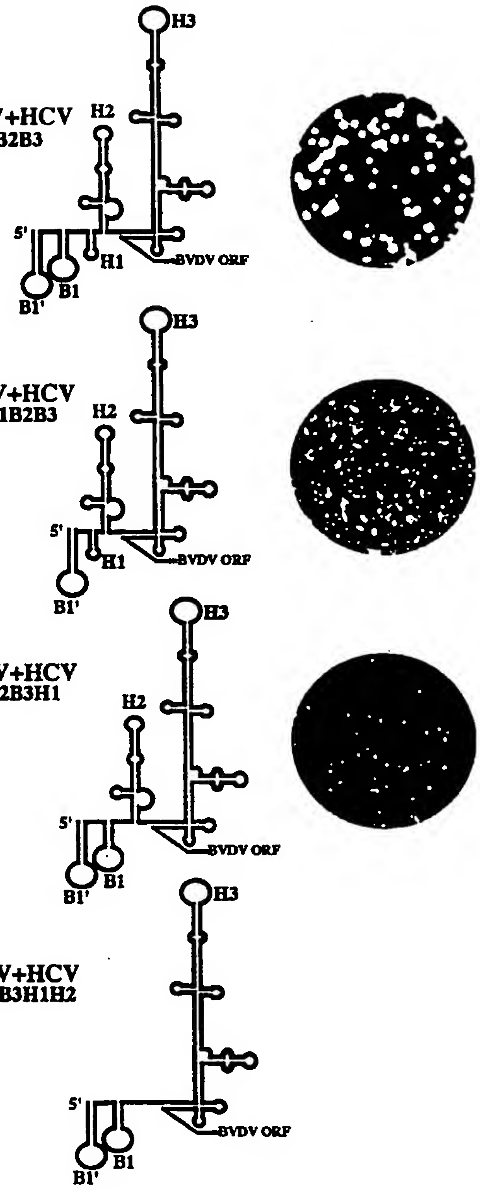

EMCV
A
B
C
D
F
G
poly C
H
I_a
I_b
I_c
J
K
L
AUG
5'
3'
HCV
H1
H2
H3
AUG
5'
3'
BVDV
B1
B1'
B2
B3
AUG
5'
3'

FIGURE 1



	Translation efficiency (% of BVDV)	Infectivity (PFU/μg of RNA)	Virus titer (PFU/ml) 24 h	48 h	Pseudorevertants
BVDV	100	$4x10^6$	$1x10^8$	$8x10^7$	No
5'HCV	50	30-50	<50	$4x10^5$ (72 h)	Yes
BVDV+HCV	25	$5x10^6$	$1x10^8$	$1x10^8$	Yes
BVDV+HCV delB3	16	$4x10^6$	$7x10^7$	$1x10^8$	N.D.

FIGURE 2A



	Translation efficiency (% of BVDV)	Infectivity (PFU/µg of RNA)	Virus titer (PFU/ml) 24h	48h	Pseudorevertants
BVDV+HCV delB2B3	24	3x10^6	7x10^7	1x10^8	N.D.
BVDV+HCV delB1B2B3	29	3x10^6	1x10^7	8x10^7	Yes
BVDV+HCV delB2B3H1	24	2x10^6	2x10^7	1x10^8	N.D.
BVDV+HCV delB2B3H1H2	3	0	<50	<50	N.D.

FIGURE 2B

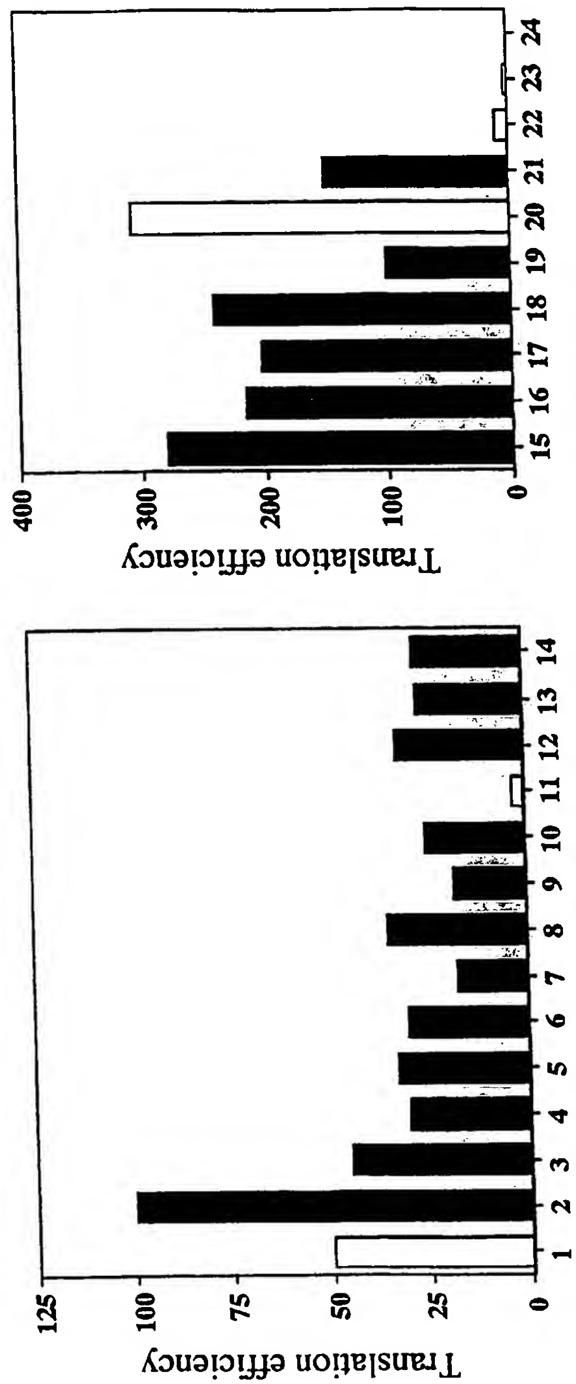
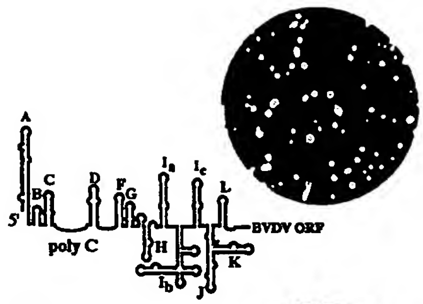
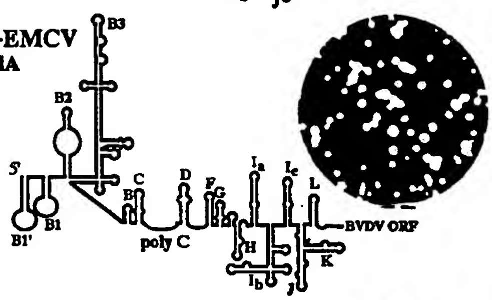

Translation efficiency
0
100
200
300
400
15 16 17 18 19 20 21 22 23 24
Translation efficiency
0
25
50
75
100
125
1 2 3 4 5 6 7 8 9 10 11 12 13 14

FIGURE 3

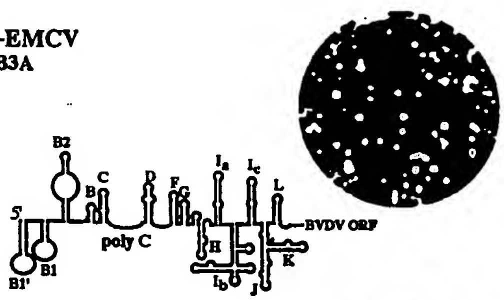
5'EMCV



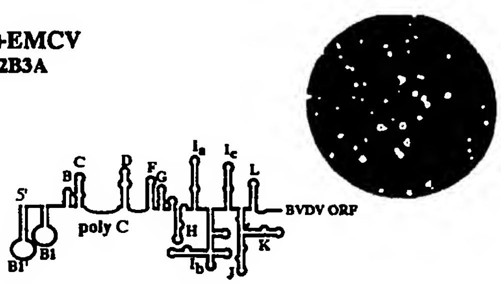
BVDV+EMCV
delA



BVDV+EMCV
delB3A



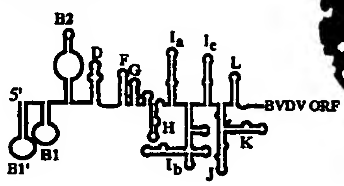
BVDV+EMCV
delB2B3A

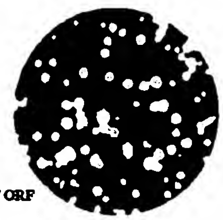


Construct	Translation efficiency (% of BVDV)	Infectivity (PFU/µg of RNA)	Virus titer (PFU/ml) 24h	Virus titer (PFU/ml) 48h	Pseudorevertants
5'EMCV	307	$2x10^3$	$5x10^2$	$3x10^6$ (72h)	Yes
BVDV+EMCV delA	153	$3x10^6$	$6x10^6$	$6x10^7$	Yes
BVDV+EMCV delB3A	280	$2x10^6$	$5x10^5$	$7x10^7$	Yes
BVDV+EMCV delB2B3A	216	$4x10^6$	$6x10^5$	$5x10^7$	Yes

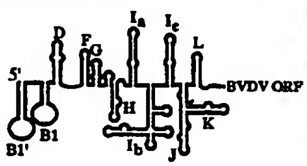
FIGURE 4A

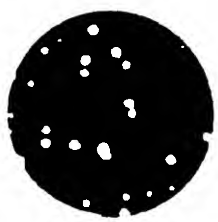
BVDV+EMCV delB3ABC



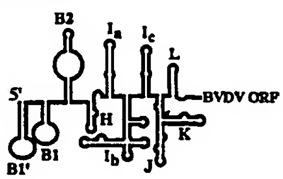


BVDV+EMCV delB2B3ABC

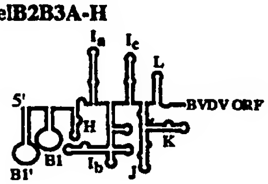




BVDV+EMCV delB3A-H



BVDV+EMCV delB2B3A-H



Construct	Translation efficiency (% of BVDV)	Infectivity (PFU/μg of RNA)	Virus titer (PFU/ml) 24h	Virus titer (PFU/ml) 48h	Pseudorevertants
BVDV+EMCV delB3ABC	202	$3x10^6$	$8x10^6$	$6x10^7$	Yes
BVDV+EMCV delB2B3ABC	240	$1x10^6$	$1x10^6$	$7x10^7$	Yes
BVDV+EMCV delB3A-H	12	0	<50	<50	No
BVDV+EMCV delB2B3A-H	3	0	<50	<50	No

FIGURE 4B

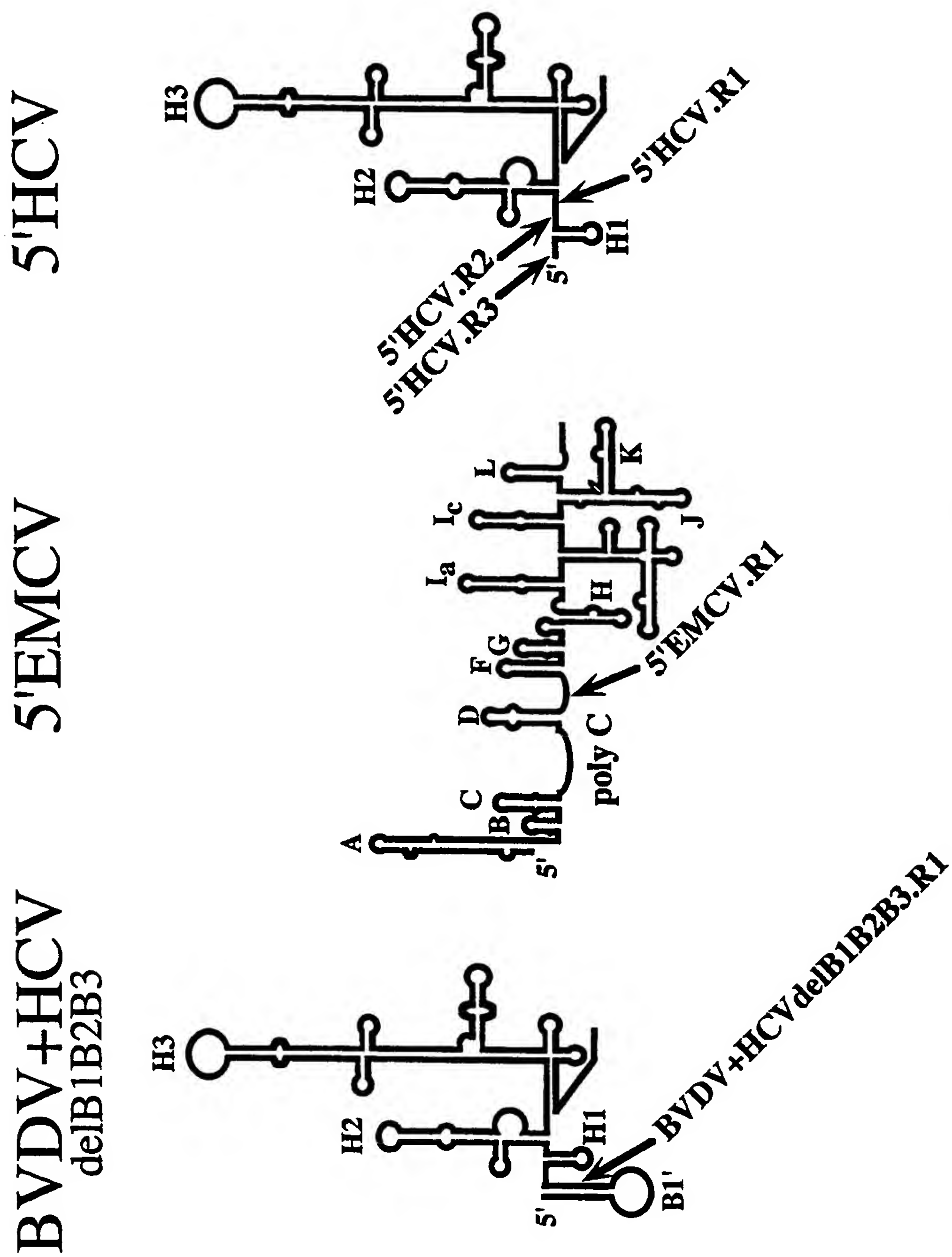

BVDV+HCV
delB1B2B3
H3
H2
H1
5'
B1'
BVDV+HCVdelB1B2B3.R1
5'EMCV
A
B
C
5'
poly C
D
F
G
H
I_a
I_c
J
L
K
5'EMCV.R1
5'HCV
H3
H2
H1
5'
5'HCV.R1
5'HCV.R2
5'HCV.R3

FIGURE 5A

B

BVDV+HCVdelB1B2B3

```
                                   <                    B1'                   >                         <       H1       >
BVDV+HCVdelB1B2B3        guauacgagaauuagaaaaggcacucguauacguaCAUGGCACGUgccagcccccugauggggc
BVDV+HCVdelB1B2B3.R1                                     guauacguaCAUGGCACGUgccagcccccugauggggc
```

5'HCV

```
                                      <       H1       >                          <       H2
5'HCV                                 gccagcccccugauggggcgacacuccaccaugaaucacuccccugugaggaac
5'HCV.R1                                                              (G/A)Uaaucacuccccugugaggaac
5'HCV.R2                     (G/A)UAUCAGAAGUGCGAAUGCUGAacacuccaccaugaaucacuccccugugaggaac
5'HCV.R3          (G/A)UAUUGCAGUUUgccagcccccugauggggcgacacuccaccaugaaucacuccccugugaggaac
```

5'EMCV

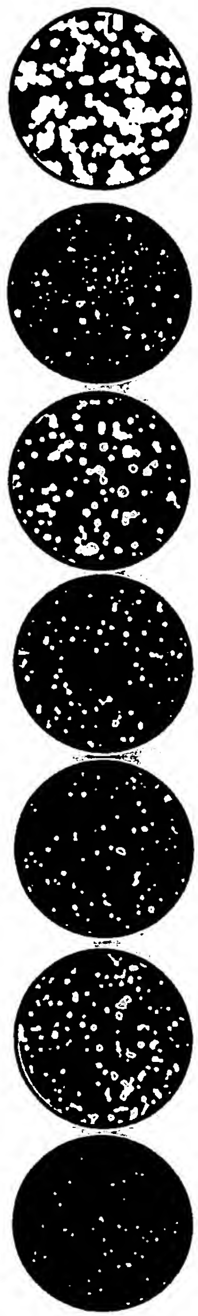
```
                 1                                             201
5'EMCV           guugaaagccgggggugggg··················  ··guuugucuauauguuauuuuccaccauauug
5'EMCV.R1                                                           Guauguuauuuuccaccauauug
```

FIGURE 5B

A

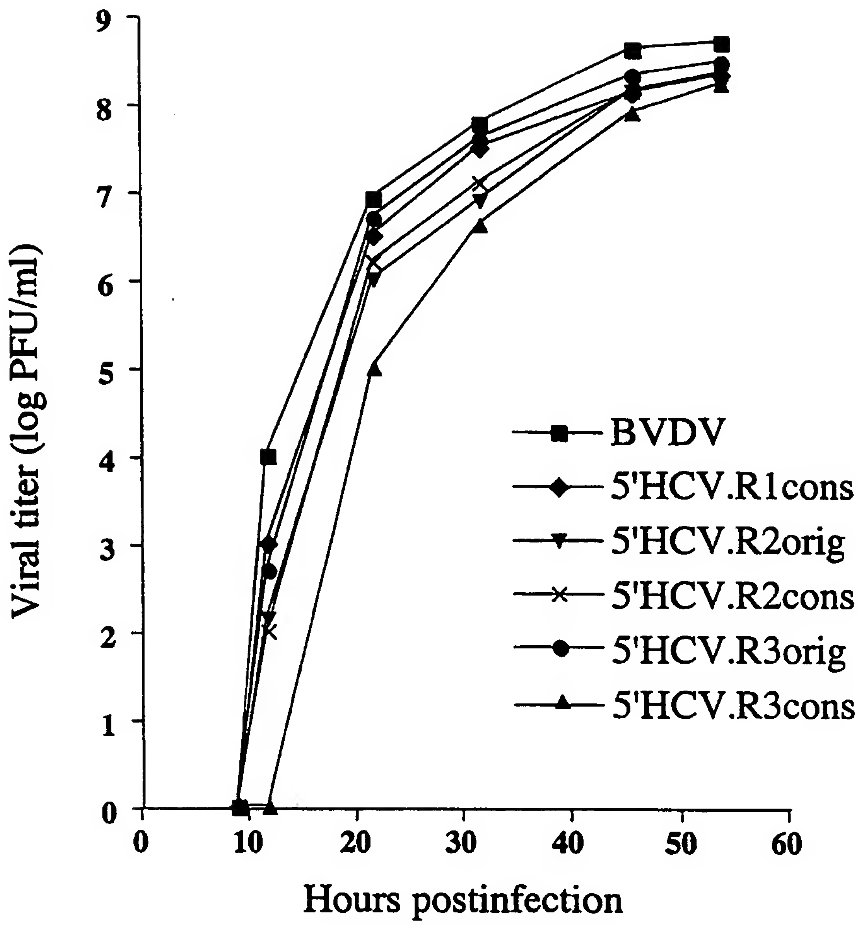
```
5'HCV                           gccagcccccugauggggcgacacuccaccaugaaucacuccccugugaggaacu
5'HCV.R1orig                                                      GUaaucacuccccugugaggaacu
5'HCV.R1cons                                                    GUAUaaucacuccccugugaggaacu
5'HCV.R2orig              GUAUCAGAAGUGCGAAUGCUGAacacuccaccaugaaucacuccccugugaggaacu
5'HCV.R2cons                                      GUAUacacuccaccaugaaucacuccccugugaggaacu
5'HCV.R3orig     GUAUUGCAGUUUgccagcccccugauggggcgacacuccaccaugaaucacuccccugugaggaacu
5'HCV.R3cons             GUAUgccagcccccugauggggcgacacuccaccaugaaucacuccccugugaggaacu
```

FIGURE 6A



	Translation efficiency (% of BVDV)	Infectivity (PFU/μg of RNA)	Virus titer (PFU/ml)	
			24h	48h
BVDV	100	$4x10^6$	$7x10^7$	$1x10^8$
5'HCV.R1orig (5'-GUAA)	45	$4x10^5$	$2x10^3$	$2x10^5$
5'HCV.R1cons (5'-GUAU)	29	$3x10^6$	$4x10^7$	$5x10^7$
5'HCV.R2orig (5'-GUAUCAGAAGUGCGAAUGCUGA)	17	$2x10^6$	$7x10^6$	$5x10^7$
5'HCV.R2cons (5'-GUAU)	35	$3x10^6$	$2x10^7$	$4x10^7$
5'HCV.R3orig (5'-GUAUUGCAGUUU)	33	$3x10^6$	$4x10^7$	$5x10^7$
5'HCV.R3cons (5'-GUAU)	30	$3x10^6$	$1x10^7$	$6x10^7$

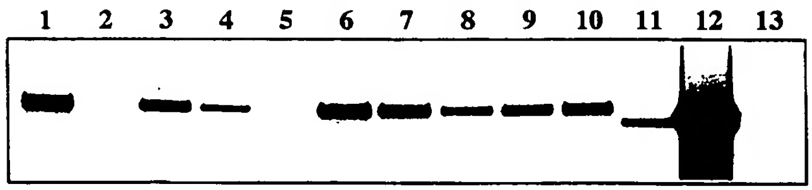
FIGURE 6B



9
8
7
6
5
4
3
2
1
0
0
10
20
30
40
50
60
Viral titer (log PFU/ml)
Hours postinfection
BVDV
5'HCV.R1cons
5'HCV.R2orig
5'HCV.R2cons
5'HCV.R3orig
5'HCV.R3cons

FIGURE 7

A


1
2
3
4
5
6
7
8
9
10
11
12
13

B

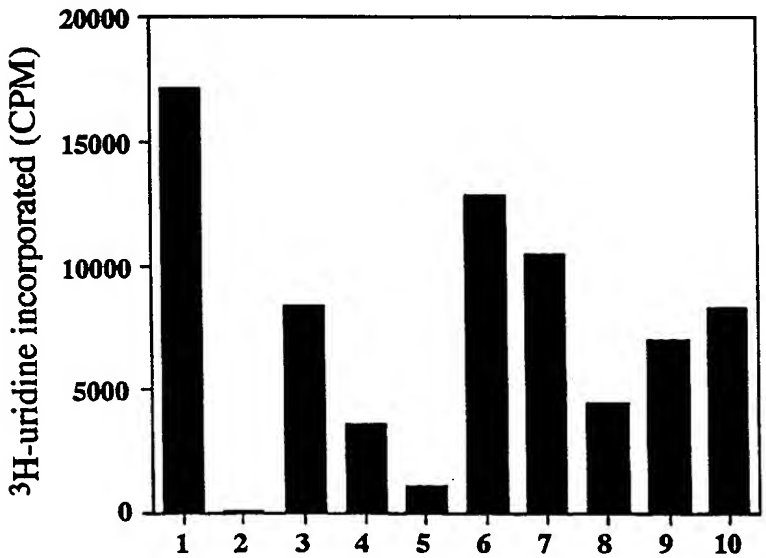

3H-uridine incorporated (CPM)
20000
15000
10000
5000
0
1
2
3
4
5
6
7
8
9
10

FIGURE 8

pACNR/BVD NADL-Xba* -> Graphic Map

DNA sequence 15065 bp gtatacgagaat ... cgactcactata circular

pACNR/BVD NADL-Xba = HaeII and XhoI digest of pACNR/BVD NADL ligated to
HaeII and XhoI digest of pACNR1180/DraIII-/BVD5'
8/27 corrected nt 12136 G to C to give HpaI site.

Co

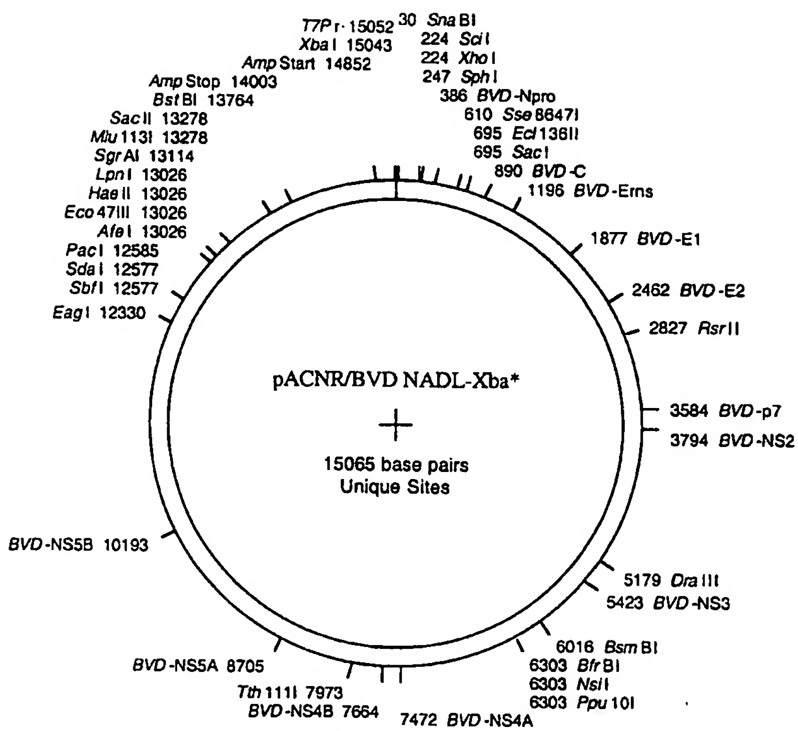


FIGURE 9

pACNR/BVD NADL-Xba* -> Genes

DNA sequence 15065 b.p. gtatacgagaat ... cgactcactata circular

pACNR/BVD NADL-Xba = HaeII and XhoI digest of pACNR/BVD NADL ligated to
HaeII and XhoI digest of pACNR1180/DraIII-/BVD5'
8/27 corrected nt 12136 G to C to give HpaI site.

Co

```
   1 gtatacgagaattagaaaaggcactcgtatacgtattgggcaattaaaaataataattaggcctagggaacaaatccctc 80

  81 tcagcgaaggccgaaaagaggctagccatgcccttagtaggactagcataatgagggggtagcaacagtggtgagttcg 160

 161 ttggatggcttaagccctgagtacagggtagtcgtcagtggttcgacgccttggaataaaggtctcgagatgccacgtgg 240

 241 acgagggcatgcccaaagcacatcttaacctgagcgggggtcgcccaggtaaaagcagttttaaccgactgttacgaata 320

 321 cagcctgatagggtgctgcagaggcccactgtattgctactaaaaatctctgctgtacatggcac ATG GAG TTG  394
   1                                                                   M   E   L    3

 395 ATC ACA AAT GAA CTT TTA TAC AAA ACA TAC AAA CAA AAA CCC GTC GGG GTG GAG GAA CCT  454
   4 I   T   N   E   L   L   Y   K   T   Y   K   Q   K   P   V   G   V   E   E   P    23

 455 GTT TAT GAT CAG GCA GGT GAT CCC TTA TTT GGT GAA AGG GGA GCA GTC CAC CCT CAA TCG  514
  24 V   Y   D   Q   A   G   D   P   L   F   G   E   R   G   A   V   H   P   Q   S    43

 515 ACG CTA AAG CTC CCA CAC AAG AGA GGG GAA CGC GAT GTT CCA ACC AAC TTG GCA TCC TTA  574
  44 T   L   K   L   P   H   K   R   G   E   R   D   V   P   T   N   L   A   S   L    63

 575 CCA AAA AGA GGT GAC TGC AGG TCG GGT AAT AGC AGA GGA CCT GTG AGC GGG ATC TAC CTG  634
  64 P   K   R   G   D   C   R   S   G   N   S   R   G   P   V   S   G   I   Y   L    83

 635 AAG CCA GGG CCA CTA TTT TAC CAG GAC TAT AAA GGT CCC GTC TAT CAC AGG GCC CCG CTG  694
  84 K   P   G   P   L   F   Y   Q   D   Y   K   G   P   V   Y   H   R   A   P   L    103

 695 GAG CTC TTT GAG GAG GGA TCC ATG TGT GAA ACG ACT AAA CGG ATA GGG AGA GTA ACT GGA  754
 104 E   L   F   E   E   G   S   M   C   E   T   T   K   R   I   G   R   V   T   G    123

 755 AGT GAC GGA AAG CTG TAC CAC ATT TAT GTG TGT ATA GAT GGA TGT ATA ATA ATA AAA AGT  814
 124 S   D   G   K   L   Y   H   I   Y   V   C   I   D   G   C   I   I   I   K   S    143

 815 GCC ACG AGA AGT TAC CAA AGG GTG TTC AGG TGG GTC CAT AAT AGG CTT GAC TGC CCT CTA  874
 144 A   T   R   S   Y   Q   R   V   F   R   W   V   H   N   R   L   D   C   P   L    163

 875 TGG GTC ACA ACT TGC TCA GAC ACG AAA GAA GAG GGA GCA ACA AAA AAG AAA ACA CAG AAA  934
 164 W   V   T   T   C   S   D   T   K   E   E   G   A   T   K   K   K   T   Q   K    183

 935 CCC GAC AGA CTA GAA AGG GGG AAA ATG AAA ATA GTG CCC AAA GAA TCT GAA AAA GAC AGC  994
 184 P   D   R   L   E   R   G   K   M   K   I   V   P   K   E   S   E   K   D   S    203

 995 AAA ACT AAA CCT CCG GAT GCT ACA ATA GTG GTG GAA GGA GTC AAA TAC CAG GTG AGG AAG  1054
 204 K   T   K   P   P   D   A   T   I   V   V   E   G   V   K   Y   Q   V   R   K    223

1055 AAG GGA AAA ACC AAG AGT AAA AAC ACT CAG GAC GGC TTG TAC CAT AAC AAA AAC AAA CCT  1114
 224 K   G   K   T   K   S   K   N   T   Q   D   G   L   Y   H   N   K   N   K   P    243

1115 CAG GAA TCA CGC AAG AAA CTG GAA AAA GCA TTG TTG GCG TGG GCA ATA ATA GCT ATA GTT  1174
 244 Q   E   S   R   K   K   L   E   K   A   L   L   A   W   A   I   I   A   I   V    263

1175 TTG TTT CAA GTT ACA ATG GGA GAA AAC ATA ACA CAG TGG AAC CTA CAA GAT AAT GGG ACG  1234
 264 L   F   Q   V   T   M   G   E   N   I   T   Q   W   N   L   Q   D   N   G   T    283

1235 GAA GGG ATA CAA CGG GCA ATG TTC CAA AGG GGT GTG AAT AGA AGT TTA CAT GGA ATC TGG  1294
 284 E   G   I   Q   R   A   M   F   Q   R   G   V   N   R   S   L   H   G   I   W    303

1295 CCA GAG AAA ATC TGT ACT GGT GTC CCT TCC CAT CTA GCC ACC GAT ATA GAA CTA AAA ACA  1354
 304 P   E   K   I   C   T   G   V   P   S   H   L   A   T   D   I   E   L   K   T    323

1355 ATT CAT GGT ATG ATG GAT GCA AGT GAG AAG ACC AAC TAC ACG TGT TGC AGA CTT CAA CGC  1414
 324 I   H   G   M   M   D   A   S   E   K   T   N   Y   T   C   C   R   L   Q   R    343

1415 CAT GAG TGG AAC AAG CAT GGT TGG TGC AAC TGG TAC AAT ATT GAA CCC TGG ATT CTA GTC  1474
 344 H   E   W   N   K   H   G   W   C   N   W   Y   N   I   E   P   W   I   L   V    363

1475 ATG AAT AGA ACC CAA GCC AAT CTC ACT GAG GGA CAA CCA CCA AGG GAG TGC GCA GTC ACT  1534
 364 M   N   R   T   Q   A   N   L   T   E   G   Q   P   P   R   E   C   A   V   T    383

1535 TGT AGG TAT GAT AGG GCT AGT GAC TTA AAC GTG GTA ACA CAA GCT AGA GAT AGC CCC ACA  1594
 384 C   R   Y   D   R   A   S   D   L   N   V   V   T   Q   A   R   D   S   P   T    403

1595 CCC TTA ACA GGT TGC AAG AAA GGA AAG AAC TTC TCC TTT GCA GGC ATA TTG ATG CGG GGC  1654
 404 P   L   T   G   C   K   K   G   K   N   F   S   F   A   G   I   L   M   R   G    423
```

FIGURE 10-1

1655 CCC TGC AAC TTT GAA ATA GCT GCA AGT GAT GTA TTA TTC AAA GAA CAT GAA CGC ATT AGT 1714
424 P C N F E I A A S D V L F K E H E R I S 443

1715 ATG TTC CAG GAT ACT ACT CTT TAC CTT GTT GAC GGG TTG ACC AAC TCC TTA GAA GGT GCC 1774
444 M F Q D T T L Y L V D G L T N S L E G A 463

1775 AGA CAA GGA ACC GCT AAA CTG ACA ACC TGG TTA GGC AAG CAG CTC GGG ATA CTA GGA AAA 1834
464 R Q G T A K L T T W L G K Q L G I L G K 483

1835 AAG TTG GAA AAC AAG AGT AAG ACG TGG TTT GGA GCA TAC GCT GCT TCC CCT TAC TGT GAT 1894
484 K L E N K S K T W F G A Y A A S P Y C D 503

1895 GTC GAT CGC AAA ATT GGC TAC ATA TGG TAT ACA AAA AAT TGC ACC CCT GCC TGC TTA CCC 1954
504 V D R K I G Y I W Y T K N C T P A C L P 523

1955 AAG AAC ACA AAA ATT GTC GGC CCT GGG AAA TTT GAC ACC AAT GCA GAG GAC GGC AAG ATA 2014
524 K N T K I V G P G K F D T N A E D G K I 543

2015 TTA CAT GAG ATG GGG GGT CAC TTG TCG GAG GTA CTA CTA CTT TCT TTA GTG GTG CTG TCC 2074
544 L H E M G G H L S E V L L L S L V V L S 563

2075 GAC TTC GCA CCG GAA ACA GCT AGT GTA ATG TAC CTA ATC CTA CAT TTT TCC ATC CCA CAA 2134
564 D F A P E T A S V M Y L I L H F S I P Q 583

2135 AGT CAC GTT GAT GTA ATG GAT TGT GAT AAG ACC CAG TTG AAC CTC ACA GTG GAG CTG ACA 2194
584 S H V D V M D C D K T Q L N L T V E L T 603

2195 ACA GCT GAA GTA ATA CCA GGG TCG GTC TGG AAT CTA GGC AAA TAT GTA TGT ATA AGA CCA 2254
604 T A E V I P G S V W N L G K Y V C I R P 623

2255 AAT TGG TGG CCT TAT GAG ACA ACT GTA GTG TTG GCA TTT GAA GAG GTG AGC CAG GTG GTG 2314
624 N W W P Y E T T V V L A F E E V S Q V V 643

2315 AAG TTA GTG TTG AGG GCA CTC AGA GAT TTA ACA CGC ATT TGG AAC GCT GCA ACA ACT ACT 2374
644 K L V L R A L R D L T R I W N A A T T T 663

2375 GCT TTT TTA GTA TGC CTT GTT AAG ATA GTC AGG GGC CAG ATG GTA CAG GGC ATT CTG TGG 2434
664 A F L V C L V K I V R G Q M V Q G I L W 683

2435 CTA CTA TTG ATA ACA GGG GTA CAA GGG CAC TTG GAT TGC AAA CCT GAA TTC TCG TAT GCC 2494
684 L L L I T G V Q G H L D C K P E F S Y A 703

2495 ATA GCA AAG GAC GAA AGA ATT GGT CAA CTG GGG GCT GAA GGC CTT ACC ACC ACT TGG AAG 2554
704 I A K D E R I G Q L G A E G L T T T W K 723

2555 GAA TAC TCA CCT GGA ATG AAG CTG GAA GAC ACA ATG GTC ATT GCT TGG TGC GAA GAT GGG 2614
724 E Y S P G M K L E D T M V I A W C E D G 743

2615 AAG TTA ATG TAC CTC CAA AGA TGC ACG AGA GAA ACC AGG TAT CTC GCA ATC TTG CAT ACA 2674
744 K L M Y L Q R C T R E T R Y L A I L H T 763

2675 AGA GCC TTG CCG ACC AGT GTG GTA TTC AAA AAA CTC TTT GAT GGG CGA AAG CAA GAG GAT 2734
764 R A L P T S V V F K K L F D G R K Q E D 783

2735 GTA GTC GAA ATG AAC GAC AAC TTT GAA TTT GGA CTC TGC CCA TGT GAT GCC AAA CCC ATA 2794
784 V V E M N D N F E F G L C P C D A K P I 803

2795 GTA AGA GGG AAG TTC AAT ACA ACG CTG CTG AAC GGA CCG GCC TTC CAG ATG GTA TGC CCC 2854
804 V R G K F N T T L L N G P A F Q M V C P 823

2855 ATA GGA TGG ACA GGG ACT GTA AGC TGT ACG TCA TTC AAT ATG GAC ACC TTA GCC ACA ACT 2914
824 I G W T G T V S C T S F N M D T L A T T 843

2915 GTG GTA CGG ACA TAT AGA AGG TCT AAA CCA TTC CCT CAT AGG CAA GGC TGT ATC ACC CAA 2974
844 V V R T Y R R S K P F P H R Q G C I T Q 863

2975 AAG AAT CTG GGG GAG GAT CTC CAT AAC TGC ATC CTT GGA GGA AAT TGG ACT TGT GTG CCT 3034
864 K N L G E D L H N C I L G G N W T C V P 883

3035 GGA GAC CAA CTA CTA TAC AAA GGG GGC TCT ATT GAA TCT TGC AAG TGG TGT GGC TAT CAA 3094
884 G D Q L L Y K G G S I E S C K W C G Y Q 903

3095 TTT AAA GAG AGT GAG GGA CTA CCA CAC TAC CCC ATT GGC AAG TGT AAA TTG GAG AAC GAG 3154
904 F K E S E G L P H Y P I G K C K L E N E 923

3155 ACT GGT TAC AGG CTA GTA GAC AGT ACC TCT TGC AAT AGA GAA GGT GTG GCC ATA GTA CCA 3214
924 T G Y R L V D S T S C N R E G V A I V P 943

3215 CAA GGG ACA TTA AAG TGC AAG ATA GGA AAA ACA ACT GTA CAG GTC ATA GCT ATG GAT ACC 3274
944 Q G T L K C K I G K T T V Q V I A M D T 963

3275 AAA CTC GGA CCT ATG CCT TGC AGA CCA TAT GAA ATC ATA TCA AGT GAG GGG CCT GTA GAA 3334
964 K L G P M P C R P Y E I I S S E G P V E 983

3335 AAG ACA GCG TGT ACT TTC AAC TAC ACT AAG ACA TTA AAA AAT AAG TAT TTT GAG CCC AGA 3394
984 K T A C T F N Y T K T L K N K Y F E P R 1003

FIGURE 10-2

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3395 GAC AGC TAC TTT CAG CAA TAC ATG CTA AAA GGA GAG TAT CAA TAC TGG TTT GAC CTG GAG 3454
1004 D   S   Y   F   Q   Q   Y   M   L   K   G   E   Y   Q   Y   W   F   D   L   E   1023

3455 GTG ACT GAC CAT CAC CGG GAT TAC TTC GCT GAG TCC ATA TTA GTG GTG GTA GTA GCC CTC 3514
1024 V   T   D   H   H   R   D   Y   F   A   E   S   I   L   V   V   V   V   A   L   1043

3515 TTG GGT GGC AGA TAT GTA CTT TGG TTA CTG GTT ACA TAC ATG GTC TTA TCA GAA CAG AAG 3574
1044 L   G   G   R   Y   V   L   W   L   L   V   T   Y   M   V   L   S   E   Q   K   1063

3575 GCC TTA GGG ATT CAG TAT GGA TCA GGG GAA GTG GTG ATG ATG GGC AAC TTG CTA ACC CAT 3634
1064 A   L   G   I   Q   Y   G   S   G   E   V   V   M   M   G   N   L   L   T   H   1083

3635 AAC AAT ATT GAA GTG GTG ACA TAC TTC TTG CTG CTG TAC CTA CTG CTG AGG GAG GAG AGC 3694
1084 N   N   I   E   V   V   T   Y   F   L   L   L   Y   L   L   L   R   E   E   S   1103

3695 GTA AAG AAG TGG GTC TTA CTC TTA TAC CAC ATC TTA GTG GTA CAC CCA ATC AAA TCT GTA 3754
1104 V   K   K   W   V   L   L   L   Y   H   I   L   V   V   H   P   I   K   S   V   1123

3755 ATT GTG ATC CTA CTG ATG ATT GGG GAT GTG GTA AAG GCC GAT TCA GGG GGC CAA GAG TAC 3814
1124 I   V   I   L   L   M   I   G   D   V   V   K   A   D   S   G   G   Q   E   Y   1143

3815 TTG GGG AAA ATA GAC CTC TGT TTT ACA ACA GTA GTA CTA ATC GTC ATA GGT TTA ATC ATA 3874
1144 L   G   K   I   D   L   C   F   T   T   V   V   L   I   V   I   G   L   I   I   1163

3875 GCC AGG CGT GAC CCA ACT ATA GTG CCA CTG GTA ACA ATA ATG GCA GCA CTG AGG GTC ACT 3934
1164 A   R   R   D   P   T   I   V   P   L   V   T   I   M   A   A   L   R   V   T   1183

3935 GAA CTG ACC CAC CAG CCT GGA GTT GAC ATC GCT GTG GCG GTC ATG ACT ATA ACC CTA CTG 3994
1184 E   L   T   H   Q   P   G   V   D   I   A   V   A   V   M   T   I   T   L   L   1203

3995 ATG GTT AGC TAT GTG ACA GAT TAT TTT AGA TAT AAA AAA TGG TTA CAG TGC ATT CTC AGC 4054
1204 M   V   S   Y   V   T   D   Y   F   R   Y   K   K   W   L   Q   C   I   L   S   1223

4055 CTG GTA TCT GCG GTG TTC TTG ATA AGA AGC CTA ATA TAC CTA GGT AGA ATC GAG ATG CCA 4114
1224 L   V   S   A   V   F   L   I   R   S   L   I   Y   L   G   R   I   E   M   P   1243

4115 GAG GTA ACT ATC CCA AAC TGG AGA CCA CTA ACT TTA ATA CTA TTA TAT TTG ATC TCA ACA 4174
1244 E   V   T   I   P   N   W   R   P   L   T   L   I   L   L   Y   L   I   S   T   1263

4175 ACA ATT GTA ACG AGG TGG AAG GTT GAC GTG GCT GGC CTA TTG TTG CAA TGT GTG CCT ATC 4234
1264 T   I   V   T   R   W   K   V   D   V   A   G   L   L   L   Q   C   V   P   I   1283

4235 TTA TTG CTG GTC ACA ACC TTG TGG GCC GAC TTC TTA ACC CTA ATA CTG ATC CTG CCT ACC 4294
1284 L   L   L   V   T   T   L   W   A   D   F   L   T   L   I   L   I   L   P   T   1303

4295 TAT GAA TTG GTT AAA TTA TAC TAT CTG AAA ACT GTT AGG ACT GAT ATA GAA AGA AGT TGG 4354
1304 Y   E   L   V   K   L   Y   Y   L   K   T   V   R   T   D   I   E   R   S   W   1323

4355 CTA GGG GGG ATA GAC TAT ACA AGA GTT GAC TCC ATC TAC GAC GTT GAT GAG AGT GGA GAG 4414
1324 L   G   G   I   D   Y   T   R   V   D   S   I   Y   D   V   D   E   S   G   E   1343

4415 GGC GTA TAT CTT TTT CCA TCA AGG CAG AAA GCA CAG GGG AAT TTT TCT ATA CTC TTG CCC 4474
1344 G   V   Y   L   F   P   S   R   Q   K   A   Q   G   N   F   S   I   L   L   P   1363

4475 CTT ATC AAA GCA ACA CTG ATA AGT TGC GTC AGC AGT AAA TGG CAG CTA ATA TAC ATG AGT 4534
1364 L   I   K   A   T   L   I   S   C   V   S   S   K   W   Q   L   I   Y   M   S   1383

4535 TAC TTA ACT TTG GAC TTT ATG TAC TAC ATG CAC AGG AAA GTT ATA GAA GAG ATC TCA GGA 4594
1384 Y   L   T   L   D   F   M   Y   Y   M   H   R   K   V   I   E   E   I   S   G   1403

4595 GGT ACC AAC ATA ATA TCC AGG TTA GTG GCA GCA CTC ATA GAG CTG AAC TGG TCC ATG GAA 4654
1404 G   T   N   I   I   S   R   L   V   A   A   L   I   E   L   N   W   S   M   E   1423

4655 GAA GAG GAG AGC AAA GGC TTA AAG AAG TTT TAT CTA TTG TCT GGA AGG TTG AGA AAC CTA 4714
1424 E   E   E   S   K   G   L   K   K   F   Y   L   L   S   G   R   L   R   N   L   1443

4715 ATA ATA AAA CAT AAG GTA AGG AAT GAG ACC GTG GCT TCT TGG TAC GGG GAG GAG GAA GTC 4774
1444 I   I   K   H   K   V   R   N   E   T   V   A   S   W   Y   G   E   E   E   V   1463

4775 TAC GGT ATG CCA AAG ATC ATG ACT ATA ATC AAG GCC AGT ACA CTG AGT AAG AGC AGG CAC 4834
1464 Y   G   M   P   K   I   M   T   I   I   K   A   S   T   L   S   K   S   R   H   1483

4835 TGC ATA ATA TGC ACT GTA TGT GAG GGC CGA GAG TGG AAA GGT GGC ACC TGC CCA AAA TGT 4894
1484 C   I   I   C   T   V   C   E   G   R   E   W   K   G   G   T   C   P   K   C   1503

4895 GGA CGC CAT GGG AAG CCG ATA ACG TGT GGG ATG TCG CTA GCA GAT TTT GAA GAA AGA CAC 4954
1504 G   R   H   G   K   P   I   T   C   G   M   S   L   A   D   F   E   E   R   H   1523

4955 TAT AAA AGA ATC TTT ATA AGG GAA GGC AAC TTT GAG GGT ATG TGC AGC CGA TGC CAG GGA 5014
1524 Y   K   R   I   F   I   R   E   G   N   F   E   G   M   C   S   R   C   Q   G   1543

5015 AAG CAT AGG AGG TTT GAA ATG GAC CGG GAA CCT AAG AGT GCC AGA TAC TGT GCT GAG TGT 5074
1544 K   H   R   R   F   E   M   D   R   E   P   K   S   A   R   Y   C   A   E   C   1563

5075 AAT AGG CTG CAT CCT GCT GAG GAA GGT GAC TTT TGG GCA GAG TCG AGC ATG TTG GGC CTC 5134
1564 N   R   L   H   P   A   E   E   G   D   F   W   A   E   S   S   M   L   G   L   1583
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FIGURE 10-3

5135 AAA ATC ACC TAC TTT GCG CTG ATG GAT GGA AAG GTG TAT GAT ATC ACA GAG TGG GCT GGA 5194
1584 K I T Y F A L M D G K V Y D I T E W A G 1603

5195 TGC CAG CGT GTG GGA ATC TCC CCA GAT ACC CAC AGA GTC CCT TGT CAC ATC TCA TTT GGT 5254
1604 C Q R V G I S P D T H R V P C H I S F G 1623

5255 TCA CGG ATG CCT TTC AGG CAG GAA TAC AAT GGC TTT GTA CAA TAT ACC GCT AGG GGG CAA 5314
1624 S R M P F R Q E Y N G F V Q Y T A R G Q 1643

5315 CTA TTT CTG AGA AAC TTG CCC GTA CTG GCA ACT AAA GTA AAA ATG CTC ATG GTA GGC AAC 5374
1644 L F L R N L P V L A T K V K M L M V G N 1663

5375 CTT GGA GAA GAA ATT GGT AAT CTG GAA CAT CTT GGG TGG ATC CTA AGG GGG CCT GCC GTG 5434
1664 L G E E I G N L E H L G W I L R G P A V 1683

5435 TGT AAG AAG ATC ACA GAG CAC GAA AAA TGC CAC ATT AAT ATA CTG GAT AAA CTA ACC GCA 5494
1684 C K K I T E H E K C H I N I L D K L T A 1703

5495 TTT TTC GGG ATC ATG CCA AGG GGG ACT ACA CCC AGA GCC CCG GTG AGG TTC CCT ACG AGC 5554
1704 F F G I M P R G T T P R A P V R F P T S 1723

5555 TTA CTA AAA GTG AGG AGG GGT CTG GAG ACT GCC TGG GCT TAC ACA CAC CAA GGC GGG ATA 5614
1724 L L K V R R G L E T A W A Y T H Q G G I 1743

5615 AGT TCA GTC GAC CAT GTA ACC GCC GGA AAA GAT CTA CTG GTC TGT GAC AGC ATG GGA CGA 5674
1744 S S V D H V T A G K D L L V C D S M G R 1763

5675 ACT AGA GTG GTT TGC CAA AGC AAC AAC AGG TTG ACC GAT GAG ACA GAG TAT GGC GTC AAG 5734
1764 T R V V C Q S N N R L T D E T E Y G V K 1783

5735 ACT GAC TCA GGG TGC CCA GAC GGT GCC AGA TGT TAT GTG TTA AAT CCA GAG GCC GTT AAC 5794
1784 T D S G C P D G A R C Y V L N P E A V N 1803

5795 ATA TCA GGA TCC AAA GGG GCA GTC GTT CAC CTC CAA AAG ACA GGT GGA GAA TTC ACG TGT 5854
1804 I S G S K G A V V H L Q K T G G E F T C 1823

5855 GTC ACC GCA TCA GGC ACA CCG GCT TTC TTC GAC CTA AAA AAC TTG AAA GGA TGG TCA GGC 5914
1824 V T A S G T P A F F D L K N L K G W S G 1843

5915 TTG CCT ATA TTT GAA GCC TCC AGC GGG AGG GTG GTT GGC AGA GTC AAA GTA GGG AAG AAT 5974
1844 L P I F E A S S G R V V G R V K V G K N 1863

5975 GAA GAG TCT AAA CCT ACA AAA ATA ATG AGT GGA ATC CAG ACC GTC TCA AAA AAC AGA GCA 6034
1864 E E S K P T K I M S G I Q T V S K N R A 1883

6035 GAC CTG ACC GAG ATG GTC AAG AAG ATA ACC AGC ATG AAC AGG GGA GAC TTC AAG CAG ATT 6094
1884 D L T E M V K K I T S M N R G D F K Q I 1903

6095 ACT TTG GCA ACA GGG GCA GGC AAA ACC ACA GAA CTC CCA AAA GCA GTT ATA GAG GAG ATA 6154
1904 T L A T G A G K T T E L P K A V I E E I 1923

6155 GGA AGA CAC AAG AGA GTA TTA GTT CTT ATA CCA TTA AGG GCA GCG GCA GAG TCA GTC TAC 6214
1924 G R H K R V L V L I P L R A A A E S V Y 1943

6215 CAG TAT ATG AGA TTG AAA CAC CCA AGC ATC TCT TTT AAC CTA AGG ATA GGG GAC ATG AAA 6274
1944 Q Y M R L K H P S I S F N L R I G D M K 1963

6275 GAG GGG GAC ATG GCA ACC GGG ATA ACC TAT GCA TCA TAC GGG TAC TTC TGC CAA ATG CCT 6334
1964 E G D M A T G I T Y A S Y G Y F C Q M P 1983

6335 CAA CCA AAG CTC AGA GCT GCT ATG GTA GAA TAC TCA TAC ATA TTC TTA GAT GAA TAC CAT 6394
1984 Q P K L R A A M V E Y S Y I F L D E Y H 2003

6395 TGT GCC ACT CCT GAA CAA CTG GCA ATT ATC GGG AAG ATC CAC AGA TTT TCA GAG AGT ATA 6454
2004 C A T P E Q L A I I G K I H R F S E S I 2023

6455 AGG GTT GTC GCC ATG ACT GCC ACG CCA GCA GGG TCG GTG ACC ACA ACA GGT CAA AAG CAC 6514
2024 R V V A M T A T P A G S V T T T G Q K H 2043

6515 CCA ATA GAG GAA TTC ATA GCC CCC GAG GTA ATG AAA GGG GAG GAT CTT GGT AGT CAG TTC 6574
2044 P I E E F I A P E V M K G E D L G S Q F 2063

6575 CTT GAT ATA GCA GGG TTA AAA ATA CCA GTG GAT GAG ATG AAA GGC AAT ATG TTG GTT TTT 6634
2064 L D I A G L K I P V D E M K G N M L V F 2083

6635 GTA CCA ACG AGA AAC ATG GCA GTA GAG GTA GCA AAG AAG CTA AAA GCT AAG GGC TAT AAC 6694
2084 V P T R N M A V E V A K K L K A K G Y N 2103

6695 TCT GGA TAC TAT TAC AGT GGA GAG GAT CCA GCC AAT CTG AGA GTT GTG ACA TCA CAA TCC 6754
2104 S G Y Y Y S G E D P A N L R V V T S Q S 2123

6755 CCC TAT GTA ATC GTG GCT ACA AAT GCT ATT GAA TCA GGA GTG ACA CTA CCA GAT TTG GAC 6814
2124 P Y V I V A T N A I E S G V T L P D L D 2143

6815 ACG GTT ATA GAC ACG GGG TTG AAA TGT GAA AAG AGG GTG AGG GTA TCA TCA AAG ATA CCC 6874
2144 T V I D T G L K C E K R V R V S S K I P 2163

FIGURE 10-4

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6875 TTC ATC GTA ACA GGC CTT AAG AGG ATG GCC GTG ACT GTG GGT GAG CAG GCG CAG CGT AGG  6934
2164 F   I   V   T   G   L   K   R   M   A   V   T   V   G   E   Q   A   Q   R   R    2183

6935 GGC AGA GTA GGT AGA GTG AAA CCC GGG AGG TAT TAT AGG AGC CAG GAA ACA GCA ACA GGG  6994
2184 G   R   V   G   R   V   K   P   G   R   Y   Y   R   S   Q   E   T   A   T   G    2203

6995 TCA AAG GAC TAC CAC TAT GAC CTC TTG CAG GCA CAA AGA TAC GGG ATT GAG GAT GGA ATC  7054
2204 S   K   D   Y   H   Y   D   L   L   Q   A   Q   R   Y   G   I   E   D   G   I    2223

7055 AAC GTG ACG AAA TCC TTT AGG GAG ATG AAT TAC GAT TGG AGC CTA TAC GAG GAG GAC AGC  7114
2224 N   V   T   K   S   F   R   E   M   N   Y   D   W   S   L   Y   E   E   D   S    2243

7115 CTA CTA ATA ACC CAG CTG GAA ATA CTA AAT AAT CTA CTC ATC TCA GAA GAC TTG CCA GCC  7174
2244 L   L   I   T   Q   L   E   I   L   N   N   L   L   I   S   E   D   L   P   A    2263

7175 GCT GTT AAG AAC ATA ATG GCC AGG ACT GAT CAC CCA GAG CCA ATC CAA CTT GCA TAC AAC  7234
2264 A   V   K   N   I   M   A   R   T   D   H   P   E   P   I   Q   L   A   Y   N    2283

7235 AGC TAT GAA GTC CAG GTC CCG GTC CTG TTC CCA AAA ATA AGG AAT GGA GAA GTC ACA GAC  7294
2284 S   Y   E   V   Q   V   P   V   L   F   P   K   I   R   N   G   E   V   T   D    2303

7295 ACC TAC GAA AAT TAC TCG TTT CTA AAT GCC AGA AAG TTA GGG GAG GAT GTG CCC GTG TAT  7354
2304 T   Y   E   N   Y   S   F   L   N   A   R   K   L   G   E   D   V   P   V   Y    2323

7355 ATC TAC GCT ACT GAA GAT GAG GAT CTG GCA GTT GAC CTC TTA GGG CTA GAC TGG CCT GAT  7414
2324 I   Y   A   T   E   D   E   D   L   A   V   D   L   L   G   L   D   W   P   D    2343

7415 CCT GGG AAC CAG CAG GTA GTG GAG ACT GGT AAA GCA CTG AAG CAA GTG ACC GGG TTG TCC  7474
2344 P   G   N   Q   Q   V   V   E   T   G   K   A   L   K   Q   V   T   G   L   S    2363

7475 TCG GCT GAA AAT GCC CTA CTA GTG GCT TTA TTT GGG TAT GTG GGT TAC CAG GCT CTC TCA  7534
2364 S   A   E   N   A   L   L   V   A   L   F   G   Y   V   G   Y   Q   A   L   S    2383

7535 AAG AGG CAT GTC CCA ATG ATA ACA GAC ATA TAT ACC ATC GAG GAC CAG AGA CTA GAA GAC  7594
2384 K   R   H   V   P   M   I   T   D   I   Y   T   I   E   D   Q   R   L   E   D    2403

7595 ACC ACC CAC CTC CAG TAT GCA CCC AAC GCC ATA AAA ACC GAT GGG ACA GAG ACT GAA CTG  7654
2404 T   T   H   L   Q   Y   A   P   N   A   I   K   T   D   G   T   E   T   E   L    2423

7655 AAA GAA CTG GCG TCG GGT GAC GTG GAA AAA ATC ATG GGA GCC ATT TCA GAT TAT GCA GCT  7714
2424 K   E   L   A   S   G   D   V   E   K   I   M   G   A   I   S   D   Y   A   A    2443

7715 GGG GGA CTG GAG TTT GTT AAA TCC CAA GCA GAA AAG ATA AAA ACA GCT CCT TTG TTT AAA  7774
2444 G   G   L   E   F   V   K   S   Q   A   E   K   I   K   T   A   P   L   F   K    2463

7775 GAA AAC GCA GAA GCC GCA AAA GGG TAT GTC CAA AAA TTC ATT GAC TCA TTA ATT GAA AAT  7834
2464 E   N   A   E   A   A   K   G   Y   V   Q   K   F   I   D   S   L   I   E   N    2483

7835 AAA GAA GAA ATA ATC AGA TAT GGT TTG TGG GGA ACA CAC ACA GCA CTA TAC AAA AGC ATA  7894
2484 K   E   E   I   I   R   Y   G   L   W   G   T   H   T   A   L   Y   K   S   I    2503

7895 GCT GCA AGA CTG GGG CAT GAA ACA GCG TTT GCC ACA CTA GTG TTA AAG TGG CTA GCT TTT  7954
2504 A   A   R   L   G   H   E   T   A   F   A   T   L   V   L   K   W   L   A   F    2523

7955 GGA GGG GAA TCA GTG TCA GAC CAC GTC AAG CAG GCG GCA GTT GAT TTA GTG GTC TAT TAT  8014
2524 G   G   E   S   V   S   D   H   V   K   Q   A   A   V   D   L   V   V   Y   Y    2543

8015 GTG ATG AAT AAG CCT TCC TTC CCA GGT GAC TCC GAG ACA CAG CAA GAA GGG AGG CGA TTC  8074
2544 V   M   N   K   P   S   F   P   G   D   S   E   T   Q   Q   E   G   R   R   F    2563

8075 GTC GCA AGC CTG TTC ATC TCC GCA CTG GCA ACC TAC ACA TAC AAA ACT TGG AAT TAC CAC  8134
2564 V   A   S   L   F   I   S   A   L   A   T   Y   T   Y   K   T   W   N   Y   H    2583

8135 AAT CTC TCT AAA GTG GTG GAA CCA GCC CTG GCT TAC CTC CCC TAT GCT ACC AGC GCA TTA  8194
2584 N   L   S   K   V   V   E   P   A   L   A   Y   L   P   Y   A   T   S   A   L    2603

8195 AAA ATG TTC ACC CCA ACG CGG CTG GAG AGC GTG GTG ATA CTG AGC ACC ACG ATA TAT AAA  8254
2604 K   M   F   T   P   T   R   L   E   S   V   V   I   L   S   T   T   I   Y   K    2623

8255 ACA TAC CTC TCT ATA AGG AAG GGG AAG AGT GAT GGA TTG CTG GGT ACG GGG ATA AGT GCA  8314
2624 T   Y   L   S   I   R   K   G   K   S   D   G   L   L   G   T   G   I   S   A    2643

8315 GCC ATG GAA ATC CTG TCA CAA AAC CCA GTA TCG GTA GGT ATA TCT GTG ATG TTG GGG GTA  8374
2644 A   M   E   I   L   S   Q   N   P   V   S   V   G   I   S   V   M   L   G   V    2663

8375 GGG GCA ATC GCT GCG CAC AAC GCT ATT GAG TCC AGT GAA CAG AAA AGG ACC CTA CTT ATG  8434
2664 G   A   I   A   A   H   N   A   I   E   S   S   E   Q   K   R   T   L   L   M    2683

8435 AAG GTG TTT GTA AAG AAC TTC TTG GAT CAG GCT GCA ACA GAT GAG CTG GTA AAA GAA AAC  8494
2684 K   V   F   V   K   N   F   L   D   Q   A   A   T   D   E   L   V   K   E   N    2703

8495 CCA GAA AAA ATT ATA ATG GCC TTA TTT GAA GCA GTC CAG ACA ATT GGT AAC CCC CTG AGA  8554
2704 P   E   K   I   I   M   A   L   F   E   A   V   Q   T   I   G   N   P   L   R    2723

8555 CTA ATA TAC CAC CTG TAT GGG GTT TAC TAC AAA GGT TGG GAG GCC AAG GAA CTA TCT GAG  8614
2724 L   I   Y   H   L   Y   G   V   Y   Y   K   G   W   E   A   K   E   L   S   E    2743
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FIGURE 10-5

8615 AGG ACA GCA GGC AGA AAC TTA TTC ACA TTG ATA ATG TTT GAA GCC TTC GAG TTA TTA GGG 8674
2744 R T A G R N L F T L I M F E A F E L L G 2763

8675 ATG GAC TCA CAA GGG AAA ATA AGG AAC CTG TCC GGA AAT TAC ATT TTG GAT TTG ATA TAC 8734
2764 M D S Q G K I R N L S G N Y I L D L I Y 2783

8735 GGC CTA CAC AAG CAA ATC AAC AGA GGG CTG AAG AAA ATG GTA CTG GGG TGG GCC CCT GCA 8794
2784 G L H K Q I N R G L K K M V L G W A P A 2803

8795 CCC TTT AGT TGT GAC TGG ACC CCT AGT GAC GAG AGG ATC AGA TTG CCA ACA GAC AAC TAT 8854
2804 P F S C D W T P S D E R I R L P T D N Y 2823

8855 TTG AGG GTA GAA ACC AGG TGC CCA TGT GGC TAT GAG ATG AAA GCT TTC AAA AAT GTA GGT 8914
2824 L R V E T R C P C G Y E M K A F K N V G 2843

8915 GGC AAA CTT ACC AAA GTG GAG GAG AGC GGG CCT TTC CTA TGT AGA AAC AGA CCT GGT AGG 8974
2844 G K L T K V E E S G P F L C R N R P G R 2863

8975 GGA CCA GTC AAC TAC AGA GTC ACC AAG TAT TAC GAT GAC AAC CTC AGA GAG ATA AAA CCA 9034
2864 G P V N Y R V T K Y Y D D N L R E I K P 2883

9035 GTA GCA AAG TTG GAA GGA CAG GTA GAG CAC TAC TAC AAA GGG GTC ACA GCA AAA ATT GAC 9094
2884 V A K L E G Q V E H Y Y K G V T A K I D 2903

9095 TAC AGT AAA GGA AAA ATG CTC TTG GCC ACT GAC AAG TGG GAG GTG GAA CAT GGT GTC ATA 9154
2904 Y S K G K M L L A T D K W E V E H G V I 2923

9155 ACC AGG TTA GCT AAG AGA TAT ACT GGG GTC GGG TTC AAT GGT GCA TAC TTA GGT GAC GAG 9214
2924 T R L A K R Y T G V G F N G A Y L G D E 2943

9215 CCC AAT CAC CGT GCT CTA GTG GAG AGG GAC TGT GCA ACT ATA ACC AAA AAC ACA GTA CAG 9274
2944 P N H R A L V E R D C A T I T K N T V Q 2963

9275 TTT CTA AAA ATG AAG AAG GGG TGT GCG TTC ACC TAT GAC CTG ACC ATC TCC AAT CTG ACC 9334
2964 F L K M K K G C A F T Y D L T I S N L T 2983

9335 AGG CTC ATC GAA CTA GTA CAC AGG AAC AAT CTT GAA GAG AAG GAA ATA CCC ACC GCT ACG 9394
2984 R L I E L V H R N N L E E K E I P T A T 3003

9395 GTC ACC ACA TGG CTA GCT TAC ACC TTC GTG AAT GAA GAC GTA GGG ACT ATA AAA CCA GTA 9454
3004 V T T W L A Y T F V N E D V G T I K P V 3023

9455 CTA GGA GAG AGA GTA ATC CCC GAC CCT GTA GTT GAT ATC AAT TTA CAA CCA GAG GTG CAA 9514
3024 L G E R V I P D P V V D I N L Q P E V Q 3043

9515 GTG GAC ACG TCA GAG GTT GGG ATC ACA ATA ATT GGA AGG GAA ACC CTG ATG ACA ACG GGA 9574
3044 V D T S E V G I T I I G R E T L M T T G 3063

9575 GTG ACA CCT GTC TTG GAA AAA GTA GAG CCT GAC GCC AGC GAC AAC CAA AAC TCG GTG AAG 9634
3064 V T P V L E K V E P D A S D N Q N S V K 3083

9635 ATC GGG TTG GAT GAG GGT AAT TAC CCA GGG CCT GGA ATA CAG ACA CAT ACA CTA ACA GAA 9694
3084 I G L D E G N Y P G P G I Q T H T L T E 3103

9695 GAA ATA CAC AAC AGG GAT GCG AGG CCC TTC ATC ATG ATC CTG GGC TCA AGG AAT TCC ATA 9754
3104 E I H N R D A R P F I M I L G S R N S I 3123

9755 TCA AAT AGG GCA AAG ACT GCT AGA AAT ATA AAT CTG TAC ACA GGA AAT GAC CCC AGG GAA 9814
3124 S N R A K T A R N I N L Y T G N D P R E 3143

9815 ATA CGA GAC TTG ATG GCT GCA GGG CGC ATG TTA GTA GTA GCA CTG AGG GAT GTC GAC CCT 9874
3144 I R D L M A A G R M L V V A L R D V D P 3163

9875 GAG CTG TCT GAA ATG GTC GAT TTC AAG GGG ACT TTT TTA GAT AGG GAG GCC CTG GAG GCT 9934
3164 E L S E M V D F K G T F L D R E A L E A 3183

9935 CTA AGT CTC GGG CAA CCT AAA CCG AAG CAG GTT ACC AAG GAA GCT GTT AGG AAT TTG ATA 9994
3184 L S L G Q P K P K Q V T K E A V R N L I 3203

9995 GAA CAG AAA AAA GAT GTG GAG ATC CCT AAC TGG TTT GCA TCA GAT GAC CCA GTA TTT CTG 10054
3204 E Q K K D V E I P N W F A S D D P V F L 3223

10055 GAA GTG GCC TTA AAA AAT GAT AAG TAC TAC TTA GTA GGA GAT GTT GGA GAG CTA AAA GAT 10114
3224 E V A L K N D K Y Y L V G D V G E L K D 3243

10115 CAA GCT AAA GCA CTT GGG GCC ACG GAT CAG ACA AGA ATT ATA AAG GAG GTA GGC TCA AGG 10174
3244 Q A K A L G A T D Q T R I I K E V G S R 3263

10175 ACG TAT GCC ATG AAG CTA TCT AGC TGG TTC CTC AAG GCA TCA AAC AAA CAG ATG AGT TTA 10234
3264 T Y A M K L S S W F L K A S N K Q M S L 3283

10235 ACT CCA CTG TTT GAG GAA TTG TTG CTA CGG TGC CCA CCT GCA ACT AAG AGC AAT AAG GGG 10294
3284 T P L F E E L L L R C P P A T K S N K G 3303

10295 CAC ATG GCA TCA GCT TAC CAA TTG GCA CAG GGT AAC TGG GAG CCC CTC GGT TGC GGG GTG 10354
3304 H M A S A Y Q L A Q G N W E P L G C G V 3323

FIGURE 10-6

10355 CAC CTA GGT ACA ATA CCA GCC AGA AGG GTG AAG ATA CAC CCA TAT GAA GCT TAC CTG AAG 10414
3324 H L G T I P A R R V K I H P Y E A Y L K 3343

10415 TTG AAA GAT TTC ATA GAA GAA GAA GAG AAG AAA CCT AGG GTT AAG GAT ACA GTA ATA AGA 10474
3344 L K D F I E E E E K K P R V K D T V I R 3363

10475 GAG CAC AAC AAA TGG ATA CTT AAA AAA ATA AGG TTT CAA GGA AAC CTC AAC ACC AAG AAA 10534
3364 E H N K W I L K K I R F Q G N L N T K K 3383

10535 ATG CTC AAC CCG GGG AAA CTA TCT GAA CAG TTG GAC AGG GAG GGG CGC AAG AGG AAC ATC 10594
3384 M L N P G K L S E Q L D R E G R K R N I 3403

10595 TAC AAC CAC CAG ATT GGT ACT ATA ATG TCA AGT GCA GGC ATA AGG CTG GAG AAA TTG CCA 10654
3404 Y N H Q I G T I M S S A G I R L E K L P 3423

10655 ATA GTG AGG GCC CAA ACC GAC ACC AAA ACC TTT CAT GAG GCA ATA AGA GAT AAG ATA GAC 10714
3424 I V R A Q T D T K T F H E A I R D K I D 3443

10715 AAG AGT GAA AAC CGG CAA AAT CCA GAA TTG CAC AAC AAA TTG TTG GAG ATT TTC CAC ACG 10774
3444 K S E N R Q N P E L H N K L L E I F H T 3463

10775 ATA GCC CAA CCC ACC CTG AAA CAC ACC TAC GGT GAG GTG ACG TGG GAG CAA CTT GAG GCG 10834
3464 I A Q P T L K H T Y G E V T W E Q L E A 3483

10835 GGG ATA AAT AGA AAG GGG GCA GCA GGC TTC CTG GAG AAG AAG AAC ATC GGA GAA GTA TTG 10894
3484 G I N R K G A A G F L E K K N I G E V L 3503

10895 GAT TCA GAA AAG CAC CTG GTA GAA CAA TTG GTC AGG GAT CTG AAG GCC GGG AGA AAG ATA 10954
3504 D S E K H L V E Q L V R D L K A G R K I 3523

10955 AAA TAT TAT GAA ACT GCA ATA CCA AAA AAT GAG AAG AGA GAT GTC AGT GAT GAC TGG CAG 11014
3524 K Y Y E T A I P K N E K R D V S D D W Q 3543

11015 GCA GGG GAC CTG GTG GTT GAG AAG AGG CCA AGA GTT ATC CAA TAC CCT GAA GCC AAG ACA 11074
3544 A G D L V V E K R P R V I Q Y P E A K T 3563

11075 AGG CTA GCC ATC ACT AAG GTC ATG TAT AAC TGG GTG AAA CAG CAG CCC GTT GTG ATT CCA 11134
3564 R L A I T K V M Y N W V K Q Q P V V I P 3583

11135 GGA TAT GAA GGA AAG ACC CCC TTG TTC AAC ATC TTT GAT AAA GTG AGA AAG GAA TGG GAC 11194
3584 G Y E G K T P L F N I F D K V R K E W D 3603

11195 TCG TTC AAT GAG CCA GTG GCC GTA AGT TTT GAC ACC AAA GCC TGG GAC ACT CAA GTG ACT 11254
3604 S F N E P V A V S F D T K A W D T Q V T 3623

11255 AGT AAG GAT CTG CAA CTT ATT GGA GAA ATC CAG AAA TAT TAC TAT AAG AAG GAG TGG CAC 11314
3624 S K D L Q L I G E I Q K Y Y Y K K E W H 3643

11315 AAG TTC ATT GAC ACC ATC ACC GAC CAC ATG ACA GAA GTA CCA GTT ATA ACA GCA GAT GGT 11374
3644 K F I D T I T D H M T E V P V I T A D G 3663

11375 GAA GTA TAT ATA AGA AAT GGG CAG AGA GGG AGC GGC CAG CCA GAC ACA AGT GCT GGC AAC 11434
3664 E V Y I R N G Q R G S G Q P D T S A G N 3683

11435 AGC ATG TTA AAT GTC CTG ACA ATG ATG TAC GGC TTC TGC GAA AGC ACA GGG GTA CCG TAC 11494
3684 S M L N V L T M M Y G F C E S T G V P Y 3703

11495 AAG AGT TTC AAC AGG GTG GCA AGG ATC CAC GTC TGT GGG GAT GAT GGC TTC TTA ATA ACT 11554
3704 K S F N R V A R I H V C G D D G F L I T 3723

11555 GAA AAA GGG TTA GGG CTG AAA TTT GCT AAC AAA GGG ATG CAG ATT CTT CAT GAA GCA GGC 11614
3724 E K G L G L K F A N K G M Q I L H E A G 3743

11615 AAA CCT CAG AAG ATA ACG GAA GGG GAA AAG ATG AAA GTT GCC TAT AGA TTT GAG GAT ATA 11674
3744 K P Q K I T E G E K M K V A Y R F E D I 3763

11675 GAG TTC TGT TCT CAT ACC CCA GTC CCT GTT AGG TGG TCC GAC AAC ACC AGT AGT CAC ATG 11734
3764 E F C S H T P V P V R W S D N T S S H M 3783

11735 GCC GGG AGA GAC ACC GCT GTG ATA CTA TCA AAG ATG GCA ACA AGA TTG GAT TCA AGT GGA 11794
3784 A G R D T A V I L S K M A T R L D S S G 3803

11795 GAG AGG GGT ACC ACA GCA TAT GAA AAA GCG GTA GCC TTC AGT TTC TTG CTG ATG TAT TCC 11854
3804 E R G T T A Y E K A V A F S F L L M Y S 3823

11855 TGG AAC CCG CTT GTT AGG AGG ATT TGC CTG TTG GTC CTT TCG CAA CAG CCA GAG ACA GAC 11914
3824 W N P L V R R I C L L V L S Q Q P E T D 3843

11915 CCA TCA AAA CAT GCC ACT TAT TAT TAC AAA GGT GAT CCA ATA GGG GCC TAT AAA GAT GTA 11974
3844 P S K H A T Y Y Y K G D P I G A Y K D V 3863

11975 ATA GGT CGG AAT CTA AGT GAA CTG AAG AGA ACA GGC TTT GAG AAA TTG GCA AAT CTA AAC 12034
3864 I G R N L S E L K R T G F E K L A N L N 3883

12035 CTA AGC CTG TCC ACG TTG GGG ATC TGG ACT AAG CAC ACA AGC AAA AGA ATA ATT CAG GAC 12094
3884 L S L S T L G I W T K H T S K R I I Q D 3903

FIGURE 10-7

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12095 TGT GTT GCC ATT GGG AAA GAA GAG GGC AAC TGG CTA GTT AAC GCC GAC AGG CTG ATA TCC 12154
 3904 C   V   A   I   G   K   E   E   G   N   W   L   V   N   A   D   R   L   I   S   3923

12155 AGC AAA ACT GGC CAC TTA TAC ATA CCT GAT AAA GGC TTT ACA TTA CAA GGA AAG CAT TAT 12214
 3924 S   K   T   G   H   L   Y   I   P   D   K   G   F   T   L   Q   G   K   H   Y   3943

12215 GAG CAA CTG CAG CTA AGA ACA GAG ACA AAC CCG GTC ATG GGG GTT GGG ACT GAG AGA TAC 12274
 3944 E   Q   L   Q   L   R   T   E   T   N   P   V   M   G   V   G   T   E   R   Y   3963

12275 AAG TTA GGT CCC ATA GTC AAT CTG CTG CTG AGA AGG TTG AAA ATT CTG CTC ATG ACG GCC 12334
 3964 K   L   G   P   I   V   N   L   L   L   R   R   L   K   I   L   L   M   T   A   3983

12335 GTC GGC GTC AGC AGC TGA gacaaaatgtatatattgtaaataaattaatccatgtacatagtgtatataaatat 12408
 3984 V   G   V   S   S   *                                                              3989

12409 agttgggaccgtccacctcaagaagacgacacgcccaacacgcacagctaaacagtagtcaagattatctacctcaagat 12488

12489 aacactacatttaatgcacacagcactttagctgtatgaggatacgcccgacgtctatagttggactagggaagacctct 12568

12569 aacagcccccctgcaggttaattaactagtgggaatacgcggggtatgccgcgttttagcatattgacgacccaattctca 12648

12649 tgtttgacagcttatcatcgtcgagcaagacgtttcccgttgaatatggctcataacaccccttgtattactgtttatgt 12728

12729 aagcagacagttttattgttcatgatgatatatttttatcttgtgcaatgtaacatcagagattttgagacacgtggctt 12808

12809 tgttgaataaatcgaacttttgctgagttgaaggatcagatcacgcatcttcccgacaacgcagaccgttccgtggcaaa 12888

12889 gcaaaagttcaaaatcaccaactggtccacctacaacaaagctctcatcaaccgtggctccctcactttctggctggatg 12968

12969 atggggcgattcaggcctggtatgagtcagcaacaccttcttcacgaggcagacctcagcgctagcggagtgtatactgg 13048

13049 cttactatgttggcactgatgagggtgtcagtgaagtgcttcatgtggcaggagaaaaaaggctgcaccggtgcgtcagc 13128

13129 agaatatgtgatacaggatatattccgcttcctcgctcactgactcgctacgctcggtcgttcgactgcggcgagcggaa 13208

13209 atggcttacgaacggggcggagatttcctggaagatgccaggaagatacttaacagggaagtgagagggccgcggcaaag 13288

13289 ccgtttttccataggctccgcccccctgacaagcatcacgaaatctgacgctcaaatcagtggtggcgaaacccgacagg 13368

13369 actataaagataccaggcgtttccccctggcggctccctcgtgcgctctcctgttcctgcctttcggtttaccggtgtcat 13448

13449 tccgctgttatggccgcgtttgtctcattccacgcctgacactcagttccgggtaggcagttcgctccaagctggactgt 13528

13529 atgcacgaaccccccgttcagtccgaccgctgcgccttatccggtaactatcgtcttgagtccaacccggaaagacatgc 13608

13609 aaaagcaccactggcagcagccactggtaattgatttagaggagttagtcttgaagtcatgcgccggttaaggctaaact 13688

13689 gaaaggacaagttttggtgactgcgctcctccaagccagttacctcggttcaaagagttggtagctcagagaaccttcga 13768

13769 aaaaccgccctgcaaggcggttttttcgttttcagagcaagagattacgcgcagaccaaaacgatctcaagaagatcatc 13848

13849 ttattaaggggtctgacgctcagtggaacgaaaactcacgttaagggattttggtcatgagattatcaaaaaggatcttc 13928

13929 acctagatccttttaaattaaaaatgaagttttaaatcaatctaaagtatatatgagtaaacttggtctgacagttacca 14008

14009 atgcttaatcagtgaggcacctatctcagcgatctgtctatttcgttcatccatagttgcctgactccccgtcgtgtaga 14088

14089 taactacgatacgggagggcttaccatctggccccagtgctgcaatgataccgcgagacccacgctcaccggctccagat 14168

14169 ttatcagcaataaaccagccagccggaagggccgagcgcagaagtggtcctgcaactttatccgcctccatccagtctat 14248

14249 taattgttgccgggaagctagagtaagtagttcgccagttaatagtttgcgcaacgttgttgccattgctgcaggcatcg 14328

14329 tggtgtcacgctcgtcgtttggtatggcttcattcagctccggttcccaacgatcaaggcgagttacatgatcccccatg 14408

14409 ttgtgcaaaaaagcggttagctccttcggtcctccgatcgttgtcagaagtaagttggccgcagtgttatcactcatggt 14488

14489 tatggcagcactgcataattctcttactgtcatgccatccgtaagatgcttttctgtgactggtgagtactcaaccaagt 14568

14569 cattctgagaatagtgtatgcggcgaccgagttgctcttgcccggcgtcaacacgggataataccgcgccacatagcaga 14648

14649 actttaaaagtgctcatcattggaaaacgttcttcggggcgaaaactctcaaggatcttaccgctgttgagatccagttc 14728

14729 gatgtaacccactcgtgcacccaactgatcttcagcatcttttactttcaccagcgtttctgggtgagcaaaaacaggaa 14808

14809 ggcaaaatgccgcaaaaaagggaataagggcgacacggaaatgttgaatactcatactcttcctttttcaatattattga 14888

14889 agcatttatcagggttattgtctcatgagcggatacatatttgaatgtatttagaaaaataaacaaataggggttccgcg 14968

14969 cacatttccccgaaaagtgccacctgacgtcgacctgaggtaattataacccgggccctatatatggatccaattctaga 15048

15049 taatacgactcactata                                                                15065
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FIGURE 10-8

BVDV NADL (inf. clone) -> Genes

DNA sequence 12578 b.p. gtatacgagaat ... ctaacagccccc linear

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   1 gtatacgagaattagaaaaggcactcgtatacgtattgggcaattaaaaataataattaggcctagggaacaaatccctc 80

  81 tcagcgaaggccgaaaagaggctagccatgcccttagtaggactagcataatgaggggggtagcaacagtggtgagttcg 160

 161 ttggatggcttaagccctgagtacagggtagtcgtcagtggttcgacgccttggaataaaggtctcgagatgccacgtgg 240

 241 acgagggcatgcccaaagcacatcttaacctgagcgggggtcgcccaggtaaaagcagttttaaccgactgttacgaata 320

 321 cagcctgatagggtgctgcagaggcccactgtattgctactaaaaatctctgctgtacatggcac ATG GAG TTG   394
   1                                                                   M   E   L     3

 395 ATC ACA AAT GAA CTT TTA TAC AAA ACA TAC AAA CAA AAA CCC GTC GGG GTG GAG GAA CCT  454
   4 I   T   N   E   L   L   Y   K   T   Y   K   Q   K   P   V   G   V   E   E   P    23

 455 GTT TAT GAT CAG GCA GGT GAT CCC TTA TTT GGT GAA AGG GGA GCA GTC CAC CCT CAA TCG  514
  24 V   Y   D   Q   A   G   D   P   L   F   G   E   R   G   A   V   H   P   Q   S    43

 515 ACG CTA AAG CTC CCA CAC AAG AGA GGG GAA CGC GAT GTT CCA ACC AAC TTG GCA TCC TTA  574
  44 T   L   K   L   P   H   K   R   G   E   R   D   V   P   T   N   L   A   S   L    63

 575 CCA AAA AGA GGT GAC TGC AGG TCG GGT AAT AGC AGA GGA CCT GTG AGC GGG ATC TAC CTG  634
  64 P   K   R   G   D   C   R   S   G   N   S   R   G   P   V   S   G   I   Y   L    83

 635 AAG CCA GGG CCA CTA TTT TAC CAG GAC TAT AAA GGT CCC GTC TAT CAC AGG GCC CCG CTG  694
  84 K   P   G   P   L   F   Y   Q   D   Y   K   G   P   V   Y   H   R   A   P   L   103

 695 GAG CTC TTT GAG GAG GGA TCC ATG TGT GAA ACG ACT AAA CGG ATA GGG AGA GTA ACT GGA  754
 104 E   L   F   E   E   G   S   M   C   E   T   T   K   R   I   G   R   V   T   G   123

 755 AGT GAC GGA AAG CTG TAC CAC ATT TAT GTG TGT ATA GAT GGA TGT ATA ATA ATA AAA AGT  814
 124 S   D   G   K   L   Y   H   I   Y   V   C   I   D   G   C   I   I   I   K   S   143

 815 GCC ACG AGA AGT TAC CAA AGG GTG TTC AGG TGG GTC CAT AAT AGG CTT GAC TGC CCT CTA  874
 144 A   T   R   S   Y   Q   R   V   F   R   W   V   H   N   R   L   D   C   P   L   163

 875 TGG GTC ACA ACT TGC TCA GAC ACG AAA GAA GAG GGA GCA ACA AAA AAG AAA ACA CAG AAA  934
 164 W   V   T   T   C   S   D   T   K   E   E   G   A   T   K   K   K   T   Q   K   183

 935 CCC GAC AGA CTA GAA AGG GGG AAA ATG AAA ATA GTG CCC AAA GAA TCT GAA AAA GAC AGC  994
 184 P   D   R   L   E   R   G   K   M   K   I   V   P   K   E   S   E   K   D   S   203

 995 AAA ACT AAA CCT CCG GAT GCT ACA ATA GTG GTG GAA GGA GTC AAA TAC CAG GTG AGG AAG  1054
 204 K   T   K   P   P   D   A   T   I   V   V   E   G   V   K   Y   Q   V   R   K   223

1055 AAG GGA AAA ACC AAG AGT AAA AAC ACT CAG GAC GGC TTG TAC CAT AAC AAA AAC AAA CCT  1114
 224 K   G   K   T   K   S   K   N   T   Q   D   G   L   Y   H   N   K   N   K   P   243

1115 CAG GAA TCA CGC AAG AAA CTG GAA AAA GCA TTG TTG GCG TGG GCA ATA ATA GCT ATA GTT  1174
 244 Q   E   S   R   K   K   L   E   K   A   L   L   A   W   A   I   I   A   I   V   263

1175 TTG TTT CAA GTT ACA ATG GGA GAA AAC ATA ACA CAG TGG AAC CTA CAA GAT AAT GGG ACG  1234
 264 L   F   Q   V   T   M   G   E   N   I   T   Q   W   N   L   Q   D   N   G   T   283

1235 GAA GGG ATA CAA CGG GCA ATG TTC CAA AGG GGT GTG AAT AGA AGT TTA CAT GGA ATC TGG  1294
 284 E   G   I   Q   R   A   M   F   Q   R   G   V   N   R   S   L   H   G   I   W   303

1295 CCA GAG AAA ATC TGT ACT GGT GTC CCT TCC CAT CTA GCC ACC GAT ATA GAA CTA AAA ACA  1354
 304 P   E   K   I   C   T   G   V   P   S   H   L   A   T   D   I   E   L   K   T   323

1355 ATT CAT GGT ATG ATG GAT GCA AGT GAG AAG ACC AAC TAC ACG TGT TGC AGA CTT CAA CGC  1414
 324 I   H   G   M   M   D   A   S   E   K   T   N   Y   T   C   C   R   L   Q   R   343

1415 CAT GAG TGG AAC AAG CAT GGT TGG TGC AAC TGG TAC AAT ATT GAA CCC TGG ATT CTA GTC  1474
 344 H   E   W   N   K   H   G   W   C   N   W   Y   N   I   E   P   W   I   L   V   363

1475 ATG AAT AGA ACC CAA GCC AAT CTC ACT GAG GGA CAA CCA CCA AGG GAG TGC GCA GTC ACT  1534
 364 M   N   R   T   Q   A   N   L   T   E   G   Q   P   P   R   E   C   A   V   T   383

1535 TGT AGG TAT GAT AGG GCT AGT GAC TTA AAC GTG GTA ACA CAA GCT AGA GAT AGC CCC ACA  1594
 384 C   R   Y   D   R   A   S   D   L   N   V   V   T   Q   A   R   D   S   P   T   403

1595 CCC TTA ACA GGT TGC AAG AAA GGA AAG AAC TTC TCC TTT GCA GGC ATA TTG ATG CGG GGC  1654
 404 P   L   T   G   C   K   K   G   K   N   F   S   F   A   G   I   L   M   R   G   423

1655 CCC TGC AAC TTT GAA ATA GCT GCA AGT GAT GTA TTA TTC AAA GAA CAT GAA CGC ATT AGT  1714
 424 P   C   N   F   E   I   A   A   S   D   V   L   F   K   E   H   E   R   I   S   443

1715 ATG TTC CAG GAT ACT ACT CTT TAC CTT GTT GAC GGG TTG ACC AAC TCC TTA GAA GGT GCC  1774
 444 M   F   Q   D   T   T   L   Y   L   V   D   G   L   T   N   S   L   E   G   A   463
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FIGURE 11-1

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1775 AGA CAA GGA ACC GCT AAA CTG ACA ACC TGG TTA GGC AAG CAG CTC GGG ATA CTA GGA AAA 1834
 464 R   Q   G   T   A   K   L   T   T   W   L   G   K   Q   L   G   I   L   G   K   483

1835 AAG TTG GAA AAC AAG AGT AAG ACG TGG TTT GGA GCA TAC GCT GCT TCC CCT TAC TGT GAT 1894
 484 K   L   E   N   K   S   K   T   W   F   G   A   Y   A   A   S   P   Y   C   D   503

1895 GTC GAT CGC AAA ATT GGC TAC ATA TGG TAT ACA AAA AAT TGC ACC CCT GCC TGC TTA CCC 1954
 504 V   D   R   K   I   G   Y   I   W   Y   T   K   N   C   T   P   A   C   L   P   523

1955 AAG AAC ACA AAA ATT GTC GGC CCT GGG AAA TTT GAC ACC AAT GCA GAG GAC GGC AAG ATA 2014
 524 K   N   T   K   I   V   G   P   G   K   F   D   T   N   A   E   D   G   K   I   543

2015 TTA CAT GAG ATG GGG GGT CAC TTG TCG GAG GTA CTA CTA CTT TCT TTA GTG GTG CTG TCC 2074
 544 L   H   E   M   G   G   H   L   S   E   V   L   L   L   S   L   V   V   L   S   563

2075 GAC TTC GCA CCG GAA ACA GCT AGT GTA ATG TAC CTA ATC CTA CAT TTT TCC ATC CCA CAA 2134
 564 D   F   A   P   E   T   A   S   V   M   Y   L   I   L   H   F   S   I   P   Q   583

2135 AGT CAC GTT GAT GTA ATG GAT TGT GAT AAG ACC CAG TTG AAC CTC ACA GTG GAG CTG ACA 2194
 584 S   H   V   D   V   M   D   C   D   K   T   Q   L   N   L   T   V   E   L   T   603

2195 ACA GCT GAA GTA ATA CCA GGG TCG GTC TGG AAT CTA GGC AAA TAT GTA TGT ATA AGA CCA 2254
 604 T   A   E   V   I   P   G   S   V   W   N   L   G   K   Y   V   C   I   R   P   623

2255 AAT TGG TGG CCT TAT GAG ACA ACT GTA GTG TTG GCA TTT GAA GAG GTG AGC CAG GTG GTG 2314
 624 N   W   W   P   Y   E   T   T   V   V   L   A   F   E   E   V   S   Q   V   V   643

2315 AAG TTA GTG TTG AGG GCA CTC AGA GAT TTA ACA CGC ATT TGG AAC GCT GCA ACA ACT ACT 2374
 644 K   L   V   L   R   A   L   R   D   L   T   R   I   W   N   A   A   T   T   T   663

2375 GCT TTT TTA GTA TGC CTT GTT AAG ATA GTC AGG GGC CAG ATG GTA CAG GGC ATT CTG TGG 2434
 664 A   F   L   V   C   L   V   K   I   V   R   G   Q   M   V   Q   G   I   L   W   683

2435 CTA CTA TTG ATA ACA GGG GTA CAA GGG CAC TTG GAT TGC AAA CCT GAA TTC TCG TAT GCC 2494
 684 L   L   L   I   T   G   V   Q   G   H   L   D   C   K   P   E   F   S   Y   A   703

2495 ATA GCA AAG GAC GAA AGA ATT GGT CAA CTG GGG GCT GAA GGC CTT ACC ACC ACT TGG AAG 2554
 704 I   A   K   D   E   R   I   G   Q   L   G   A   E   G   L   T   T   T   W   K   723

2555 GAA TAC TCA CCT GGA ATG AAG CTG GAA GAC ACA ATG GTC ATT GCT TGG TGC GAA GAT GGG 2614
 724 E   Y   S   P   G   M   K   L   E   D   T   M   V   I   A   W   C   E   D   G   743

2615 AAG TTA ATG TAC CTC CAA AGA TGC ACG AGA GAA ACC AGG TAT CTC GCA ATC TTG CAT ACA 2674
 744 K   L   M   Y   L   Q   R   C   T   R   E   T   R   Y   L   A   I   L   H   T   763

2675 AGA GCC TTG CCG ACC AGT GTG GTA TTC AAA AAA CTC TTT GAT GGG CGA AAG CAA GAG GAT 2734
 764 R   A   L   P   T   S   V   V   F   K   K   L   F   D   G   R   K   Q   E   D   783

2735 GTA GTC GAA ATG AAC GAC AAC TTT GAA TTT GGA CTC TGC CCA TGT GAT GCC AAA CCC ATA 2794
 784 V   V   E   M   N   D   N   F   E   F   G   L   C   P   C   D   A   K   P   I   803

2795 GTA AGA GGG AAG TTC AAT ACA ACG CTG CTG AAC GGA CCG GCC TTC CAG ATG GTA TGC CCC 2854
 804 V   R   G   K   F   N   T   T   L   L   N   G   P   A   F   Q   M   V   C   P   823

2855 ATA GGA TGG ACA GGG ACT GTA AGC TGT ACG TCA TTC AAT ATG GAC ACC TTA GCC ACA ACT 2914
 824 I   G   W   T   G   T   V   S   C   T   S   F   N   M   D   T   L   A   T   T   843

2915 GTG GTA CGG ACA TAT AGA AGG TCT AAA CCA TTC CCT CAT AGG CAA GGC TGT ATC ACC CAA 2974
 844 V   V   R   T   Y   R   R   S   K   P   F   P   H   R   Q   G   C   I   T   Q   863

2975 AAG AAT CTG GGG GAG GAT CTC CAT AAC TGC ATC CTT GGA GGA AAT TGG ACT TGT GTG CCT 3034
 864 K   N   L   G   E   D   L   H   N   C   I   L   G   G   N   W   T   C   V   P   883

3035 GGA GAC CAA CTA CTA TAC AAA GGG GGC TCT ATT GAA TCT TGC AAG TGG TGT GGC TAT CAA 3094
 884 G   D   Q   L   L   Y   K   G   G   S   I   E   S   C   K   W   C   G   Y   Q   903

3095 TTT AAA GAG AGT GAG GGA CTA CCA CAC TAC CCC ATT GGC AAG TGT AAA TTG GAG AAC GAG 3154
 904 F   K   E   S   E   G   L   P   H   Y   P   I   G   K   C   K   L   E   N   E   923

3155 ACT GGT TAC AGG CTA GTA GAC AGT ACC TCT TGC AAT AGA GAA GGT GTG GCC ATA GTA CCA 3214
 924 T   G   Y   R   L   V   D   S   T   S   C   N   R   E   G   V   A   I   V   P   943

3215 CAA GGG ACA TTA AAG TGC AAG ATA GGA AAA ACA ACT GTA CAG GTC ATA GCT ATG GAT ACC 3274
 944 Q   G   T   L   K   C   K   I   G   K   T   T   V   Q   V   I   A   M   D   T   963

3275 AAA CTC GGA CCT ATG CCT TGC AGA CCA TAT GAA ATC ATA TCA AGT GAG GGG CCT GTA GAA 3334
 964 K   L   G   P   M   P   C   R   P   Y   E   I   I   S   S   E   G   P   V   E   983

3335 AAG ACA GCG TGT ACT TTC AAC TAC ACT AAG ACA TTA AAA AAT AAG TAT TTT GAG CCC AGA 3394
 984 K   T   A   C   T   F   N   Y   T   K   T   L   K   N   K   Y   F   E   P   R  1003

3395 GAC AGC TAC TTT CAG CAA TAC ATG CTA AAA GGA GAG TAT CAA TAC TGG TTT GAC CTG GAG 3454
1004 D   S   Y   F   Q   Q   Y   M   L   K   G   E   Y   Q   Y   W   F   D   L   E  1023

3455 GTG ACT GAC CAT CAC CGG GAT TAC TTC GCT GAG TCC ATA TTA GTG GTG GTA GTA GCC CTC 3514
1024 V   T   D   H   H   R   D   Y   F   A   E   S   I   L   V   V   V   V   A   L  1043
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FIGURE 11-2

3515 TTG GGT GGC AGA TAT GTA CTT TGG TTA CTG GTT ACA TAC ATG GTC TTA TCA GAA CAG AAG 3574
1044 L G G R Y V L W L L V T Y M V L S E Q K 1063

3575 GCC TTA GGG ATT CAG TAT GGA TCA GGG GAA GTG GTG ATG ATG GGC AAC TTG CTA ACC CAT 3634
1064 A L G I Q Y G S G E V V M M G N L L T H 1083

3635 AAC AAT ATT GAA GTG GTG ACA TAC TTC TTG CTG CTG TAC CTA CTG CTG AGG GAG GAG AGC 3694
1084 N N I E V V T Y F L L L Y L L L R E E S 1103

3695 GTA AAG AAG TGG GTC TTA CTC TTA TAC CAC ATC TTA GTG GTA CAC CCA ATC AAA TCT GTA 3754
1104 V K K W V L L L Y H I L V V H P I K S V 1123

3755 ATT GTG ATC CTA CTG ATG ATT GGG GAT GTG GTA AAG GCC GAT TCA GGG GGC CAA GAG TAC 3814
1124 I V I L L M I G D V V K A D S G G Q E Y 1143

3815 TTG GGG AAA ATA GAC CTC TGT TTT ACA ACA GTA GTA CTA ATC GTC ATA GGT TTA ATC ATA 3874
1144 L G K I D L C F T T V V L I V I G L I I 1163

3875 GCC AGG CGT GAC CCA ACT ATA GTG CCA CTG GTA ACA ATA ATG GCA GCA CTG AGG GTC ACT 3934
1164 A R R D P T I V P L V T I M A A L R V T 1183

3935 GAA CTG ACC CAC CAG CCT GGA GTT GAC ATC GCT GTG GCG GTC ATG ACT ATA ACC CTA CTG 3994
1184 E L T H Q P G V D I A V A V M T I T L L 1203

3995 ATG GTT AGC TAT GTG ACA GAT TAT TTT AGA TAT AAA AAA TGG TTA CAG TGC ATT CTC AGC 4054
1204 M V S Y V T D Y F R Y K K W L Q C I L S 1223

4055 CTG GTA TCT GCG GTG TTC TTG ATA AGA AGC CTA ATA TAC CTA GGT AGA ATC GAG ATG CCA 4114
1224 L V S A V F L I R S L I Y L G R I E M P 1243

4115 GAG GTA ACT ATC CCA AAC TGG AGA CCA CTA ACT TTA ATA CTA TTA TAT TTG ATC TCA ACA 4174
1244 E V T I P N W R P L T L I L L Y L I S T 1263

4175 ACA ATT GTA ACG AGG TGG AAG GTT GAC GTG GCT GGC CTA TTG TTG CAA TGT GTG CCT ATC 4234
1264 T I V T R W K V D V A G L L L Q C V P I 1283

4235 TTA TTG CTG GTC ACA ACC TTG TGG GCC GAC TTC TTA ACC CTA ATA CTG ATC CTG CCT ACC 4294
1284 L L L V T T L W A D F L T L I L I L P T 1303

4295 TAT GAA TTG GTT AAA TTA TAC TAT CTG AAA ACT GTT AGG ACT GAT ATA GAA AGA AGT TGG 4354
1304 Y E L V K L Y Y L K T V R T D I E R S W 1323

4355 CTA GGG GGG ATA GAC TAT ACA AGA GTT GAC TCC ATC TAC GAC GTT GAT GAG AGT GGA GAG 4414
1324 L G G I D Y T R V D S I Y D V D E S G E 1343

4415 GGC GTA TAT CTT TTT CCA TCA AGG CAG AAA GCA CAG GGG AAT TTT TCT ATA CTC TTG CCC 4474
1344 G V Y L F P S R Q K A Q G N F S I L L P 1363

4475 CTT ATC AAA GCA ACA CTG ATA AGT TGC GTC AGC AGT AAA TGG CAG CTA ATA TAC ATG AGT 4534
1364 L I K A T L I S C V S S K W Q L I Y M S 1383

4535 TAC TTA ACT TTG GAC TTT ATG TAC TAC ATG CAC AGG AAA GTT ATA GAA GAG ATC TCA GGA 4594
1384 Y L T L D F M Y Y M H R K V I E E I S G 1403

4595 GGT ACC AAC ATA ATA TCC AGG TTA GTG GCA GCA CTC ATA GAG CTG AAC TGG TCC ATG GAA 4654
1404 G T N I I S R L V A A L I E L N W S M E 1423

4655 GAA GAG GAG AGC AAA GGC TTA AAG AAG TTT TAT CTA TTG TCT GGA AGG TTG AGA AAC CTA 4714
1424 E E E S K G L K K F Y L L S G R L R N L 1443

4715 ATA ATA AAA CAT AAG GTA AGG AAT GAG ACC GTG GCT TCT TGG TAC GGG GAG GAG GAA GTC 4774
1444 I I K H K V R N E T V A S W Y G E E E V 1463

4775 TAC GGT ATG CCA AAG ATC ATG ACT ATA ATC AAG GCC AGT ACA CTG AGT AAG AGC AGG CAC 4834
1464 Y G M P K I M T I I K A S T L S K S R H 1483

4835 TGC ATA ATA TGC ACT GTA TGT GAG GGC CGA GAG TGG AAA GGT GGC ACC TGC CCA AAA TGT 4894
1484 C I I C T V C E G R E W K G G T C P K C 1503

4895 GGA CGC CAT GGG AAG CCG ATA ACG TGT GGG ATG TCG CTA GCA GAT TTT GAA GAA AGA CAC 4954
1504 G R H G K P I T C G M S L A D F E E R H 1523

4955 TAT AAA AGA ATC TTT ATA AGG GAA GGC AAC TTT GAG GGT ATG TGC AGC CGA TGC CAG GGA 5014
1524 Y K R I F I R E G N F E G M C S R C Q G 1543

5015 AAG CAT AGG AGG TTT GAA ATG GAC CGG GAA CCT AAG AGT GCC AGA TAC TGT GCT GAG TGT 5074
1544 K H R R F E M D R E P K S A R Y C A E C 1563

5075 AAT AGG CTG CAT CCT GCT GAG GAA GGT GAC TTT TGG GCA GAG TCG AGC ATG TTG GGC CTC 5134
1564 N R L H P A E E G D F W A E S S M L G L 1583

5135 AAA ATC ACC TAC TTT GCG CTG ATG GAT GGA AAG GTG TAT GAT ATC ACA GAG TGG GCT GGA 5194
1584 K I T Y F A L M D G K V Y D I T E W A G 1603

5195 TGC CAG CGT GTG GGA ATC TCC CCA GAT ACC CAC AGA GTC CCT TGT CAC ATC TCA TTT GGT 5254
1604 C Q R V G I S P D T H R V P C H I S F G 1623

FIGURE 11-3

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5255 TCA CGG ATG CCT TTC AGG CAG GAA TAC AAT GGC TTT GTA CAA TAT ACC GCT AGG GGG CAA  5314
1624 S   R   M   P   F   R   Q   E   Y   N   G   F   V   Q   Y   T   A   R   G   Q    1643

5315 CTA TTT CTG AGA AAC TTG CCC GTA CTG GCA ACT AAA GTA AAA ATG CTC ATG GTA GGC AAC  5374
1644 L   F   L   R   N   L   P   V   L   A   T   K   V   K   M   L   M   V   G   N    1663

5375 CTT GGA GAA GAA ATT GGT AAT CTG GAA CAT CTT GGG TGG ATC CTA AGG GGG CCT GCC GTG  5434
1664 L   G   E   E   I   G   N   L   E   H   L   G   W   I   L   R   G   P   A   V    1683

5435 TGT AAG AAG ATC ACA GAG CAC GAA AAA TGC CAC ATT AAT ATA CTG GAT AAA CTA ACC GCA  5494
1684 C   K   K   I   T   E   H   E   K   C   H   I   N   I   L   D   K   L   T   A    1703

5495 TTT TTC GGG ATC ATG CCA AGG GGG ACT ACA CCC AGA GCC CCG GTG AGG TTC CCT ACG AGC  5554
1704 F   F   G   I   M   P   R   G   T   T   P   R   A   P   V   R   F   P   T   S    1723

5555 TTA CTA AAA GTG AGG AGG GGT CTG GAG ACT GCC TGG GCT TAC ACA CAC CAA GGC GGG ATA  5614
1724 L   L   K   V   R   R   G   L   E   T   A   W   A   Y   T   H   Q   G   G   I    1743

5615 AGT TCA GTC GAC CAT GTA ACC GCC GGA AAA GAT CTA CTG GTC TGT GAC AGC ATG GGA CGA  5674
1744 S   S   V   D   H   V   T   A   G   K   D   L   L   V   C   D   S   M   G   R    1763

5675 ACT AGA GTG GTT TGC CAA AGC AAC AAC AGG TTG ACC GAT GAG ACA GAG TAT GGC GTC AAG  5734
1764 T   R   V   V   C   Q   S   N   N   R   L   T   D   E   T   E   Y   G   V   K    1783

5735 ACT GAC TCA GGG TGC CCA GAC GGT GCC AGA TGT TAT GTG TTA AAT CCA GAG GCC GTT AAC  5794
1784 T   D   S   G   C   P   D   G   A   R   C   Y   V   L   N   P   E   A   V   N    1803

5795 ATA TCA GGA TCC AAA GGG GCA GTC GTT CAC CTC CAA AAG ACA GGT GGA GAA TTC ACG TGT  5854
1804 I   S   G   S   K   G   A   V   V   H   L   Q   K   T   G   G   E   F   T   C    1823

5855 GTC ACC GCA TCA GGC ACA CCG GCT TTC TTC GAC CTA AAA AAC TTG AAA GGA TGG TCA GGC  5914
1824 V   T   A   S   G   T   P   A   F   F   D   L   K   N   L   K   G   W   S   G    1843

5915 TTG CCT ATA TTT GAA GCC TCC AGC GGG AGG GTG GTT GGC AGA GTC AAA GTA GGG AAG AAT  5974
1844 L   P   I   F   E   A   S   S   G   R   V   V   G   R   V   K   V   G   K   N    1863

5975 GAA GAG TCT AAA CCT ACA AAA ATA ATG AGT GGA ATC CAG ACC GTC TCA AAA AAC AGA GCA  6034
1864 E   E   S   K   P   T   K   I   M   S   G   I   Q   T   V   S   K   N   R   A    1883

6035 GAC CTG ACC GAG ATG GTC AAG AAG ATA ACC AGC ATG AAC AGG GGA GAC TTC AAG CAG ATT  6094
1884 D   L   T   E   M   V   K   K   I   T   S   M   N   R   G   D   F   K   Q   I    1903

6095 ACT TTG GCA ACA GGG GCA GGC AAA ACC ACA GAA CTC CCA AAA GCA GTT ATA GAG GAG ATA  6154
1904 T   L   A   T   G   A   G   K   T   T   E   L   P   K   A   V   I   E   E   I    1923

6155 GGA AGA CAC AAG AGA GTA TTA GTT CTT ATA CCA TTA AGG GCA GCG GCA GAG TCA GTC TAC  6214
1924 G   R   H   K   R   V   L   V   L   I   P   L   R   A   A   A   E   S   V   Y    1943

6215 CAG TAT ATG AGA TTG AAA CAC CCA AGC ATC TCT TTT AAC CTA AGG ATA GGG GAC ATG AAA  6274
1944 Q   Y   M   R   L   K   H   P   S   I   S   F   N   L   R   I   G   D   M   K    1963

6275 GAG GGG GAC ATG GCA ACC GGG ATA ACC TAT GCA TCA TAC GGG TAC TTC TGC CAA ATG CCT  6334
1964 E   G   D   M   A   T   G   I   T   Y   A   S   Y   G   Y   F   C   Q   M   P    1983

6335 CAA CCA AAG CTC AGA GCT GCT ATG GTA GAA TAC TCA TAC ATA TTC TTA GAT GAA TAC CAT  6394
1984 Q   P   K   L   R   A   A   M   V   E   Y   S   Y   I   F   L   D   E   Y   H    2003

6395 TGT GCC ACT CCT GAA CAA CTG GCA ATT ATC GGG AAG ATC CAC AGA TTT TCA GAG AGT ATA  6454
2004 C   A   T   P   E   Q   L   A   I   I   G   K   I   H   R   F   S   E   S   I    2023

6455 AGG GTT GTC GCC ATG ACT GCC ACG CCA GCA GGG TCG GTG ACC ACA ACA GGT CAA AAG CAC  6514
2024 R   V   V   A   M   T   A   T   P   A   G   S   V   T   T   T   G   Q   K   H    2043

6515 CCA ATA GAG GAA TTC ATA GCC CCC GAG GTA ATG AAA GGG GAG GAT CTT GGT AGT CAG TTC  6574
2044 P   I   E   E   F   I   A   P   E   V   M   K   G   E   D   L   G   S   Q   F    2063

6575 CTT GAT ATA GCA GGG TTA AAA ATA CCA GTG GAT GAG ATG AAA GGC AAT ATG TTG GTT TTT  6634
2064 L   D   I   A   G   L   K   I   P   V   D   E   M   K   G   N   M   L   V   F    2083

6635 GTA CCA ACG AGA AAC ATG GCA GTA GAG GTA GCA AAG AAG CTA AAA GCT AAG GGC TAT AAC  6694
2084 V   P   T   R   N   M   A   V   E   V   A   K   K   L   K   A   K   G   Y   N    2103

6695 TCT GGA TAC TAT TAC AGT GGA GAG GAT CCA GCC AAT CTG AGA GTT GTG ACA TCA CAA TCC  6754
2104 S   G   Y   Y   Y   S   G   E   D   P   A   N   L   R   V   V   T   S   Q   S    2123

6755 CCC TAT GTA ATC GTG GCT ACA AAT GCT ATT GAA TCA GGA GTG ACA CTA CCA GAT TTG GAC  6814
2124 P   Y   V   I   V   A   T   N   A   I   E   S   G   V   T   L   P   D   L   D    2143

6815 ACG GTT ATA GAC ACG GGG TTG AAA TGT GAA AAG AGG GTG AGG GTA TCA TCA AAG ATA CCC  6874
2144 T   V   I   D   T   G   L   K   C   E   K   R   V   R   V   S   S   K   I   P    2163

6875 TTC ATC GTA ACA GGC CTT AAG AGG ATG GCC GTG ACT GTG GGT GAG CAG GCG CAG CGT AGG  6934
2164 F   I   V   T   G   L   K   R   M   A   V   T   V   G   E   Q   A   Q   R   R    2183

6935 GGC AGA GTA GGT AGA GTG AAA CCC GGG AGG TAT TAT AGG AGC CAG GAA ACA GCA ACA GGG  6994
2184 G   R   V   G   R   V   K   P   G   R   Y   Y   R   S   Q   E   T   A   T   G    2203
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FIGURE 11-4

6995 TCA AAG GAC TAC CAC TAT GAC CTC TTG CAG GCA CAA AGA TAC GGG ATT GAG GAT GGA ATC 7054
2204 S K D Y H Y D L L Q A Q R Y G I E D G I 2223

7055 AAC GTG ACG AAA TCC TTT AGG GAG ATG AAT TAC GAT TGG AGC CTA TAC GAG GAG GAC AGC 7114
2224 N V T K S F R E M N Y D W S L Y E E D S 2243

7115 CTA CTA ATA ACC CAG CTG GAA ATA CTA AAT AAT CTA CTC ATC TCA GAA GAC TTG CCA GCC 7174
2244 L L I T Q L E I L N N L L I S E D L P A 2263

7175 GCT GTT AAG AAC ATA ATG GCC AGG ACT GAT CAC CCA GAG CCA ATC CAA CTT GCA TAC AAC 7234
2264 A V K N I M A R T D H P E P I Q L A Y N 2283

7235 AGC TAT GAA GTC CAG GTC CCG GTC CTG TTC CCA AAA ATA AGG AAT GGA GAA GTC ACA GAC 7294
2284 S Y E V Q V P V L F P K I R N G E V T D 2303

7295 ACC TAC GAA AAT TAC TCG TTT CTA AAT GCC AGA AAG TTA GGG GAG GAT GTG CCC GTG TAT 7354
2304 T Y E N Y S F L N A R K L G E D V P V Y 2323

7355 ATC TAC GCT ACT GAA GAT GAG GAT CTG GCA GTT GAC CTC TTA GGG CTA GAC TGG CCT GAT 7414
2324 I Y A T E D E D L A V D L L G L D W P D 2343

7415 CCT GGG AAC CAG CAG GTA GTG GAG ACT GGT AAA GCA CTG AAG CAA GTG ACC GGG TTG TCC 7474
2344 P G N Q Q V V E T G K A L K Q V T G L S 2363

7475 TCG GCT GAA AAT GCC CTA CTA GTG GCT TTA TTT GGG TAT GTG GGT TAC CAG GCT CTC TCA 7534
2364 S A E N A L L V A L F G Y V G Y Q A L S 2383

7535 AAG AGG CAT GTC CCA ATG ATA ACA GAC ATA TAT ACC ATC GAG GAC CAG AGA CTA GAA GAC 7594
2384 K R H V P M I T D I Y T I E D Q R L E D 2403

7595 ACC ACC CAC CTC CAG TAT GCA CCC AAC GCC ATA AAA ACC GAT GGG ACA GAG ACT GAA CTG 7654
2404 T T H L Q Y A P N A I K T D G T E T E L 2423

7655 AAA GAA CTG GCG TCG GGT GAC GTG GAA AAA ATC ATG GGA GCC ATT TCA GAT TAT GCA GCT 7714
2424 K E L A S G D V E K I M G A I S D Y A A 2443

7715 GGG GGA CTG GAG TTT GTT AAA TCC CAA GCA GAA AAG ATA AAA ACA GCT CCT TTG TTT AAA 7774
2444 G G L E F V K S Q A E K I K T A P L F K 2463

7775 GAA AAC GCA GAA GCC GCA AAA GGG TAT GTC CAA AAA TTC ATT GAC TCA TTA ATT GAA AAT 7834
2464 E N A E A A K G Y V Q K F I D S L I E N 2483

7835 AAA GAA GAA ATA ATC AGA TAT GGT TTG TGG GGA ACA CAC ACA GCA CTA TAC AAA AGC ATA 7894
2484 K E E I I R Y G L W G T H T A L Y K S I 2503

7895 GCT GCA AGA CTG GGG CAT GAA ACA GCG TTT GCC ACA CTA GTG TTA AAG TGG CTA GCT TTT 7954
2504 A A R L G H E T A F A T L V L K W L A F 2523

7955 GGA GGG GAA TCA GTG TCA GAC CAC GTC AAG CAG GCG GCA GTT GAT TTA GTG GTC TAT TAT 8014
2524 G G E S V S D H V K Q A A V D L V V Y Y 2543

8015 GTG ATG AAT AAG CCT TCC TTC CCA GGT GAC TCC GAG ACA CAG CAA GAA GGG AGG CGA TTC 8074
2544 V M N K P S F P G D S E T Q Q E G R R F 2563

8075 GTC GCA AGC CTG TTC ATC TCC GCA CTG GCA ACC TAC ACA TAC AAA ACT TGG AAT TAC CAC 8134
2564 V A S L F I S A L A T Y T Y K T W N Y H 2583

8135 AAT CTC TCT AAA GTG GTG GAA CCA GCC CTG GCT TAC CTC CCC TAT GCT ACC AGC GCA TTA 8194
2584 N L S K V V E P A L A Y L P Y A T S A L 2603

8195 AAA ATG TTC ACC CCA ACG CGG CTG GAG AGC GTG GTG ATA CTG AGC ACC ACG ATA TAT AAA 8254
2604 K M F T P T R L E S V V I L S T T I Y K 2623

8255 ACA TAC CTC TCT ATA AGG AAG GGG AAG AGT GAT GGA TTG CTG GGT ACG GGG ATA AGT GCA 8314
2624 T Y L S I R K G K S D G L L G T G I S A 2643

8315 GCC ATG GAA ATC CTG TCA CAA AAC CCA GTA TCG GTA GGT ATA TCT GTG ATG TTG GGG GTA 8374
2644 A M E I L S Q N P V S V G I S V M L G V 2663

8375 GGG GCA ATC GCT GCG CAC AAC GCT ATT GAG TCC AGT GAA CAG AAA AGG ACC CTA CTT ATG 8434
2664 G A I A A H N A I E S S E Q K R T L L M 2683

8435 AAG GTG TTT GTA AAG AAC TTC TTG GAT CAG GCT GCA ACA GAT GAG CTG GTA AAA GAA AAC 8494
2684 K V F V K N F L D Q A A T D E L V K E N 2703

8495 CCA GAA AAA ATT ATA ATG GCC TTA TTT GAA GCA GTC CAG ACA ATT GGT AAC CCC CTG AGA 8554
2704 P E K I I M A L F E A V Q T I G N P L R 2723

8555 CTA ATA TAC CAC CTG TAT GGG GTT TAC TAC AAA GGT TGG GAG GCC AAG GAA CTA TCT GAG 8614
2724 L I Y H L Y G V Y Y K G W E A K E L S E 2743

8615 AGG ACA GCA GGC AGA AAC TTA TTC ACA TTG ATA ATG TTT GAA GCC TTC GAG TTA TTA GGG 8674
2744 R T A G R N L F T L I M F E A F E L L G 2763

8675 ATG GAC TCA CAA GGG AAA ATA AGG AAC CTG TCC GGA AAT TAC ATT TTG GAT TTG ATA TAC 8734
2764 M D S Q G K I R N L S G N Y I L D L I Y 2783

FIGURE 11-5

8735 GGC CTA CAC AAG CAA ATC AAC AGA GGG CTG AAG AAA ATG GTA CTG GGG TGG GCC CCT GCA 8794
2784 G L H K Q I N R G L K K M V L G W A P A 2803

8795 CCC TTT AGT TGT GAC TGG ACC CCT AGT GAC GAG AGG ATC AGA TTG CCA ACA GAC AAC TAT 8854
2804 P F S C D W T P S D E R I R L P T D N Y 2823

8855 TTG AGG GTA GAA ACC AGG TGC CCA TGT GGC TAT GAG ATG AAA GCT TTC AAA AAT GTA GGT 8914
2824 L R V E T R C P C G Y E M K A F K N V G 2843

8915 GGC AAA CTT ACC AAA GTG GAG GAG AGC GGG CCT TTC CTA TGT AGA AAC AGA CCT GGT AGG 8974
2844 G K L T K V E E S G P F L C R N R P G R 2863

8975 GGA CCA GTC AAC TAC AGA GTC ACC AAG TAT TAC GAT GAC AAC CTC AGA GAG ATA AAA CCA 9034
2864 G P V N Y R V T K Y Y D D N L R E I K P 2883

9035 GTA GCA AAG TTG GAA GGA CAG GTA GAG CAC TAC TAC AAA GGG GTC ACA GCA AAA ATT GAC 9094
2884 V A K L E G Q V E H Y Y K G V T A K I D 2903

9095 TAC AGT AAA GGA AAA ATG CTC TTG GCC ACT GAC AAG TGG GAG GTG GAA CAT GGT GTC ATA 9154
2904 Y S K G K M L L A T D K W E V E H G V I 2923

9155 ACC AGG TTA GCT AAG AGA TAT ACT GGG GTC GGG TTC AAT GGT GCA TAC TTA GGT GAC GAG 9214
2924 T R L A K R Y T G V G F N G A Y L G D E 2943

9215 CCC AAT CAC CGT GCT CTA GTG GAG AGG GAC TGT GCA ACT ATA ACC AAA AAC ACA GTA CAG 9274
2944 P N H R A L V E R D C A T I T K N T V Q 2963

9275 TTT CTA AAA ATG AAG AAG GGG TGT GCG TTC ACC TAT GAC CTG ACC ATC TCC AAT CTG ACC 9334
2964 F L K M K K G C A F T Y D L T I S N L T 2983

9335 AGG CTC ATC GAA CTA GTA CAC AGG AAC AAT CTT GAA GAG AAG GAA ATA CCC ACC GCT ACG 9394
2984 R L I E L V H R N N L E E K E I P T A T 3003

9395 GTC ACC ACA TGG CTA GCT TAC ACC TTC GTG AAT GAA GAC GTA GGG ACT ATA AAA CCA GTA 9454
3004 V T T W L A Y T F V N E D V G T I K P V 3023

9455 CTA GGA GAG AGA GTA ATC CCC GAC CCT GTA GTT GAT ATC AAT TTA CAA CCA GAG GTG CAA 9514
3024 L G E R V I P D P V V D I N L Q P E V Q 3043

9515 GTG GAC ACG TCA GAG GTT GGG ATC ACA ATA ATT GGA AGG GAA ACC CTG ATG ACA ACG GGA 9574
3044 V D T S E V G I T I I G R E T L M T T G 3063

9575 GTG ACA CCT GTC TTG GAA AAA GTA GAG CCT GAC GCC AGC GAC AAC CAA AAC TCG GTG AAG 9634
3064 V T P V L E K V E P D A S D N Q N S V K 3083

9635 ATC GGG TTG GAT GAG GGT AAT TAC CCA GGG CCT GGA ATA CAG ACA CAT ACA CTA ACA GAA 9694
3084 I G L D E G N Y P G P G I Q T H T L T E 3103

9695 GAA ATA CAC AAC AGG GAT GCG AGG CCC TTC ATC ATG ATC CTG GGC TCA AGG AAT TCC ATA 9754
3104 E I H N R D A R P F I M I L G S R N S I 3123

9755 TCA AAT AGG GCA AAG ACT GCT AGA AAT ATA AAT CTG TAC ACA GGA AAT GAC CCC AGG GAA 9814
3124 S N R A K T A R N I N L Y T G N D P R E 3143

9815 ATA CGA GAC TTG ATG GCT GCA GGG CGC ATG TTA GTA GTA GCA CTG AGG GAT GTC GAC CCT 9874
3144 I R D L M A A G R M L V V A L R D V D P 3163

9875 GAG CTG TCT GAA ATG GTC GAT TTC AAG GGG ACT TTT TTA GAT AGG GAG GCC CTG GAG GCT 9934
3164 E L S E M V D F K G T F L D R E A L E A 3183

9935 CTA AGT CTC GGG CAA CCT AAA CCG AAG CAG GTT ACC AAG GAA GCT GTT AGG AAT TTG ATA 9994
3184 L S L G Q P K P K Q V T K E A V R N L I 3203

9995 GAA CAG AAA AAA GAT GTG GAG ATC CCT AAC TGG TTT GCA TCA GAT GAC CCA GTA TTT CTG 10054
3204 E Q K K D V E I P N W F A S D D P V F L 3223

10055 GAA GTG GCC TTA AAA AAT GAT AAG TAC TAC TTA GTA GGA GAT GTT GGA GAG CTA AAA GAT 10114
3224 E V A L K N D K Y Y L V G D V G E L K D 3243

10115 CAA GCT AAA GCA CTT GGG GCC ACG GAT CAG ACA AGA ATT ATA AAG GAG GTA GGC TCA AGG 10174
3244 Q A K A L G A T D Q T R I I K E V G S R 3263

10175 ACG TAT GCC ATG AAG CTA TCT AGC TGG TTC CTC AAG GCA TCA AAC AAA CAG ATG AGT TTA 10234
3264 T Y A M K L S S W F L K A S N K Q M S L 3283

10235 ACT CCA CTG TTT GAG GAA TTG TTG CTA CGG TGC CCA CCT GCA ACT AAG AGC AAT AAG GGG 10294
3284 T P L F E E L L L R C P P A T K S N K G 3303

10295 CAC ATG GCA TCA GCT TAC CAA TTG GCA CAG GGT AAC TGG GAG CCC CTC GGT TGC GGG GTG 10354
3304 H M A S A Y Q L A Q G N W E P L G C G V 3323

10355 CAC CTA GGT ACA ATA CCA GCC AGA AGG GTG AAG ATA CAC CCA TAT GAA GCT TAC CTG AAG 10414
3324 H L G T I P A R R V K I H P Y E A Y L K 3343

10415 TTG AAA GAT TTC ATA GAA GAA GAA GAG AAG AAA CCT AGG GTT AAG GAT ACA GTA ATA AGA 10474
3344 L K D F I E E E E K K P R V K D T V I R 3363

FIGURE 11-6

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10475 GAG CAC AAC AAA TGG ATA CTT AAA AAA ATA AGG TTT CAA GGA AAC CTC AAC ACC AAG AAA 10534
 3364 E   H   N   K   W   I   L   K   K   I   R   F   Q   G   N   L   N   T   K   K   3383

10535 ATG CTC AAC CCG GGG AAA CTA TCT GAA CAG TTG GAC AGG GAG GGG CGC AAG AGG AAC ATC 10594
 3384 M   L   N   P   G   K   L   S   E   Q   L   D   R   E   G   R   K   R   N   I   3403

10595 TAC AAC CAC CAG ATT GGT ACT ATA ATG TCA AGT GCA GGC ATA AGG CTG GAG AAA TTG CCA 10654
 3404 Y   N   H   Q   I   G   T   I   M   S   S   A   G   I   R   L   E   K   L   P   3423

10655 ATA GTG AGG GCC CAA ACC GAC ACC AAA ACC TTT CAT GAG GCA ATA AGA GAT AAG ATA GAC 10714
 3424 I   V   R   A   Q   T   D   T   K   T   F   H   E   A   I   R   D   K   I   D   3443

10715 AAG AGT GAA AAC CGG CAA AAT CCA GAA TTG CAC AAC AAA TTG TTG GAG ATT TTC CAC ACG 10774
 3444 K   S   E   N   R   Q   N   P   E   L   H   N   K   L   L   E   I   F   H   T   3463

10775 ATA GCC CAA CCC ACC CTG AAA CAC ACC TAC GGT GAG GTG ACG TGG GAG CAA CTT GAG GCG 10834
 3464 I   A   Q   P   T   L   K   H   T   Y   G   E   V   T   W   E   Q   L   E   A   3483

10835 GGG ATA AAT AGA AAG GGG GCA GCA GGC TTC CTG GAG AAG AAG AAC ATC GGA GAA GTA TTG 10894
 3484 G   I   N   R   K   G   A   A   G   F   L   E   K   K   N   I   G   E   V   L   3503

10895 GAT TCA GAA AAG CAC CTG GTA GAA CAA TTG GTC AGG GAT CTG AAG GCC GGG AGA AAG ATA 10954
 3504 D   S   E   K   H   L   V   E   Q   L   V   R   D   L   K   A   G   R   K   I   3523

10955 AAA TAT TAT GAA ACT GCA ATA CCA AAA AAT GAG AAG AGA GAT GTC AGT GAT GAC TGG CAG 11014
 3524 K   Y   Y   E   T   A   I   P   K   N   E   K   R   D   V   S   D   D   W   Q   3543

11015 GCA GGG GAC CTG GTG GTT GAG AAG AGG CCA AGA GTT ATC CAA TAC CCT GAA GCC AAG ACA 11074
 3544 A   G   D   L   V   V   E   K   R   P   R   V   I   Q   Y   P   E   A   K   T   3563

11075 AGG CTA GCC ATC ACT AAG GTC ATG TAT AAC TGG GTG AAA CAG CAG CCC GTT GTG ATT CCA 11134
 3564 R   L   A   I   T   K   V   M   Y   N   W   V   K   Q   Q   P   V   V   I   P   3583

11135 GGA TAT GAA GGA AAG ACC CCC TTG TTC AAC ATC TTT GAT AAA GTG AGA AAG GAA TGG GAC 11194
 3584 G   Y   E   G   K   T   P   L   F   N   I   F   D   K   V   R   K   E   W   D   3603

11195 TCG TTC AAT GAG CCA GTG GCC GTA AGT TTT GAC ACC AAA GCC TGG GAC ACT CAA GTG ACT 11254
 3604 S   F   N   E   P   V   A   V   S   F   D   T   K   A   W   D   T   Q   V   T   3623

11255 AGT AAG GAT CTG CAA CTT ATT GGA GAA ATC CAG AAA TAT TAC TAT AAG AAG GAG TGG CAC 11314
 3624 S   K   D   L   Q   L   I   G   E   I   Q   K   Y   Y   Y   K   K   E   W   H   3643

11315 AAG TTC ATT GAC ACC ATC ACC GAC CAC ATG ACA GAA GTA CCA GTT ATA ACA GCA GAT GGT 11374
 3644 K   F   I   D   T   I   T   D   H   M   T   E   V   P   V   I   T   A   D   G   3663

11375 GAA GTA TAT ATA AGA AAT GGG CAG AGA GGG AGC GGC CAG CCA GAC ACA AGT GCT GGC AAC 11434
 3664 E   V   Y   I   R   N   G   Q   R   G   S   G   Q   P   D   T   S   A   G   N   3683

11435 AGC ATG TTA AAT GTC CTG ACA ATG ATG TAC GGC TTC TGC GAA AGC ACA GGG GTA CCG TAC 11494
 3684 S   M   L   N   V   L   T   M   M   Y   G   F   C   E   S   T   G   V   P   Y   3703

11495 AAG AGT TTC AAC AGG GTG GCA AGG ATC CAC GTC TGT GGG GAT GAT GGC TTC TTA ATA ACT 11554
 3704 K   S   F   N   R   V   A   R   I   H   V   C   G   D   D   G   F   L   I   T   3723

11555 GAA AAA GGG TTA GGG CTG AAA TTT GCT AAC AAA GGG ATG CAG ATT CTT CAT GAA GCA GGC 11614
 3724 E   K   G   L   G   L   K   F   A   N   K   G   M   Q   I   L   H   E   A   G   3743

11615 AAA CCT CAG AAG ATA ACG GAA GGG GAA AAG ATG AAA GTT GCC TAT AGA TTT GAG GAT ATA 11674
 3744 K   P   Q   K   I   T   E   G   E   K   M   K   V   A   Y   R   F   E   D   I   3763

11675 GAG TTC TGT TCT CAT ACC CCA GTC CCT GTT AGG TGG TCC GAC AAC ACC AGT AGT CAC ATG 11734
 3764 E   F   C   S   H   T   P   V   P   V   R   W   S   D   N   T   S   S   H   M   3783

11735 GCC GGG AGA GAC ACC GCT GTG ATA CTA TCA AAG ATG GCA ACA AGA TTG GAT TCA AGT GGA 11794
 3784 A   G   R   D   T   A   V   I   L   S   K   M   A   T   R   L   D   S   S   G   3803

11795 GAG AGG GGT ACC ACA GCA TAT GAA AAA GCG GTA GCC TTC AGT TTC TTG CTG ATG TAT TCC 11854
 3804 E   R   G   T   T   A   Y   E   K   A   V   A   F   S   F   L   L   M   Y   S   3823

11855 TGG AAC CCG CTT GTT AGG AGG ATT TGC CTG TTG GTC CTT TCG CAA CAG CCA GAG ACA GAC 11914
 3824 W   N   P   L   V   R   R   I   C   L   L   V   L   S   Q   Q   P   E   T   D   3843

11915 CCA TCA AAA CAT GCC ACT TAT TAT TAC AAA GGT GAT CCA ATA GGG GCC TAT AAA GAT GTA 11974
 3844 P   S   K   H   A   T   Y   Y   Y   K   G   D   P   I   G   A   Y   K   D   V   3863

11975 ATA GGT CGG AAT CTA AGT GAA CTG AAG AGA ACA GGC TTT GAG AAA TTG GCA AAT CTA AAC 12034
 3864 I   G   R   N   L   S   E   L   K   R   T   G   F   E   K   L   A   N   L   N   3883

12035 CTA AGC CTG TCC ACG TTG GGG ATC TGG ACT AAG CAC ACA AGC AAA AGA ATA ATT CAG GAC 12094
 3884 L   S   L   S   T   L   G   I   W   T   K   H   T   S   K   R   I   I   Q   D   3903

12095 TGT GTT GCC ATT GGG AAA GAA GAG GGC AAC TGG CTA GTT AAC GCC GAC AGG CTG ATA TCC 12154
 3904 C   V   A   I   G   K   E   E   G   N   W   L   V   N   A   D   R   L   I   S   3923

12155 AGC AAA ACT GGC CAC TTA TAC ATA CCT GAT AAA GGC TTT ACA TTA CAA GGA AAG CAT TAT 12214
 3924 S   K   T   G   H   L   Y   I   P   D   K   G   F   T   L   Q   G   K   H   Y   3943
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FIGURE 11-7

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12215 GAG CAA CTG CAG CTA AGA ACA GAG ACA AAC CCG GTC ATG GGG GTT GGG ACT GAG AGA TAC  12274
 3944 E   Q   L   Q   L   R   T   E   T   N   P   V   M   G   V   G   T   E   R   Y    3963

12275 AAG TTA GGT CCC ATA GTC AAT CTG CTG CTG AGA AGG TTG AAA ATT CTG CTC ATG ACG GCC  12334
 3964 K   L   G   P   I   V   N   L   L   L   R   R   L   K   I   L   L   M   T   A    3983

12335 GTC GGC GTC AGC AGC TGA gacaaaatgtatatattgtaaataaattaatccatgtacatagtgtatataaatat 12408
 3984 V   G   V   S   S   *                                                           3989

12409 agttgggaccgtccacctcaagaagacgacacgcccaacacgcacagctaaacagtagtcaagattatctacctcaagat 12488

12489 aacactacatttaatgcacacagcactttagctgtatgaggatacgcccgacgtctatagttggactagggaagacctct 12568

12569 aacagccccc                                                                 12578
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FIGURE 11-8

BVDV NADL cIns- (inf. clone) -> Genes

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DNA  sequence    12308 b.p.   gtatacgagaat ... ctaacagccccc   linear

   1 gtatacgagaattagaaaaggcactcgtatacgtattgggcaattaaaaataataattaggcctagggaacaaatccctc 80

  81 tcagcgaaggccgaaaagaggctagccatgcccttagtaggactagcataatgagggggtagcaacagtggtgagttcg 160

 161 ttggatggcttaagccctgagtacagggtagtcgtcagtggttcgacgccttggaataaaggtctcgagatgccacgtgg 240

 241 acgagggcatgcccaaagcacatcttaacctgagcgggggtcgcccaggtaaaagcagttttaaccgactgttacgaata 320

 321 cagcctgatagggtgctgcagaggcccactgtattgctactaaaaatctctgctgtacatggcac ATG GAG TTG   394
   1                                                                  M   E   L     3

 395 ATC ACA AAT GAA CTT TTA TAC AAA ACA TAC AAA CAA AAA CCC GTC GGG GTG GAG GAA CCT  454
   4 I   T   N   E   L   L   Y   K   T   Y   K   Q   K   P   V   G   V   E   E   P    23

 455 GTT TAT GAT CAG GCA GGT GAT CCC TTA TTT GGT GAA AGG GGA GCA GTC CAC CCT CAA TCG  514
  24 V   Y   D   Q   A   G   D   P   L   F   G   E   R   G   A   V   H   P   Q   S    43

 515 ACG CTA AAG CTC CCA CAC AAG AGA GGG GAA CGC GAT GTT CCA ACC AAC TTG GCA TCC TTA  574
  44 T   L   K   L   P   H   K   R   G   E   R   D   V   P   T   N   L   A   S   L    63

 575 CCA AAA AGA GGT GAC TGC AGG TCG GGT AAT AGC AGA GGA CCT GTG AGC GGG ATC TAC CTG  634
  64 P   K   R   G   D   C   R   S   G   N   S   R   G   P   V   S   G   I   Y   L    83

 635 AAG CCA GGG CCA CTA TTT TAC CAG GAC TAT AAA GGT CCC GTC TAT CAC AGG GCC CCG CTG  694
  84 K   P   G   P   L   F   Y   Q   D   Y   K   G   P   V   Y   H   R   A   P   L    103

 695 GAG CTC TTT GAG GAG GGA TCC ATG TGT GAA ACG ACT AAA CGG ATA GGG AGA GTA ACT GGA  754
 104 E   L   F   E   E   G   S   M   C   E   T   T   K   R   I   G   R   V   T   G    123

 755 AGT GAC GGA AAG CTG TAC CAC ATT TAT GTG TGT ATA GAT GGA TGT ATA ATA ATA AAA AGT  814
 124 S   D   G   K   L   Y   H   I   Y   V   C   I   D   G   C   I   I   I   K   S    143

 815 GCC ACG AGA AGT TAC CAA AGG GTG TTC AGG TGG GTC CAT AAT AGG CTT GAC TGC CCT CTA  874
 144 A   T   R   S   Y   Q   R   V   F   R   W   V   H   N   R   L   D   C   P   L    163

 875 TGG GTC ACA ACT TGC TCA GAC ACG AAA GAA GAG GGA GCA ACA AAA AAG AAA ACA CAG AAA  934
 164 W   V   T   T   C   S   D   T   K   E   E   G   A   T   K   K   K   T   Q   K    183

 935 CCC GAC AGA CTA GAA AGG GGG AAA ATG AAA ATA GTG CCC AAA GAA TCT GAA AAA GAC AGC  994
 184 P   D   R   L   E   R   G   K   M   K   I   V   P   K   E   S   E   K   D   S    203

 995 AAA ACT AAA CCT CCG GAT GCT ACA ATA GTG GTG GAA GGA GTC AAA TAC CAG GTG AGG AAG  1054
 204 K   T   K   P   P   D   A   T   I   V   V   E   G   V   K   Y   Q   V   R   K    223

1055 AAG GGA AAA ACC AAG AGT AAA AAC ACT CAG GAC GGC TTG TAC CAT AAC AAA AAC AAA CCT  1114
 224 K   G   K   T   K   S   K   N   T   Q   D   G   L   Y   H   N   K   N   K   P    243

1115 CAG GAA TCA CGC AAG AAA CTG GAA AAA GCA TTG TTG GCG TGG GCA ATA ATA GCT ATA GTT  1174
 244 Q   E   S   R   K   K   L   E   K   A   L   L   A   W   A   I   I   A   I   V    263

1175 TTG TTT CAA GTT ACA ATG GGA GAA AAC ATA ACA CAG TGG AAC CTA CAA GAT AAT GGG ACG  1234
 264 L   F   Q   V   T   M   G   E   N   I   T   Q   W   N   L   Q   D   N   G   T    283

1235 GAA GGG ATA CAA CGG GCA ATG TTC CAA AGG GGT GTG AAT AGA AGT TTA CAT GGA ATC TGG  1294
 284 E   G   I   Q   R   A   M   F   Q   R   G   V   N   R   S   L   H   G   I   W    303

1295 CCA GAG AAA ATC TGT ACT GGT GTC CCT TCC CAT CTA GCC ACC GAT ATA GAA CTA AAA ACA  1354
 304 P   E   K   I   C   T   G   V   P   S   H   L   A   T   D   I   E   L   K   T    323

1355 ATT CAT GGT ATG ATG GAT GCA AGT GAG AAG ACC AAC TAC ACG TGT TGC AGA CTT CAA CGC  1414
 324 I   H   G   M   M   D   A   S   E   K   T   N   Y   T   C   C   R   L   Q   R    343

1415 CAT GAG TGG AAC AAG CAT GGT TGG TGC AAC TGG TAC AAT ATT GAA CCC TGG ATT CTA GTC  1474
 344 H   E   W   N   K   H   G   W   C   N   W   Y   N   I   E   P   W   I   L   V    363

1475 ATG AAT AGA ACC CAA GCC AAT CTC ACT GAG GGA CAA CCA CCA AGG GAG TGC GCA GTC ACT  1534
 364 M   N   R   T   Q   A   N   L   T   E   G   Q   P   P   R   E   C   A   V   T    383

1535 TGT AGG TAT GAT AGG GCT AGT GAC TTA AAC GTG GTA ACA CAA GCT AGA GAT AGC CCC ACA  1594
 384 C   R   Y   D   R   A   S   D   L   N   V   V   T   Q   A   R   D   S   P   T    403

1595 CCC TTA ACA GGT TGC AAG AAA GGA AAG AAC TTC TCC TTT GCA GGC ATA TTG ATG CGG GGC  1654
 404 P   L   T   G   C   K   K   G   K   N   F   S   F   A   G   I   L   M   R   G    423

1655 CCC TGC AAC TTT GAA ATA GCT GCA AGT GAT GTA TTA TTC AAA GAA CAT GAA CGC ATT AGT  1714
 424 P   C   N   F   E   I   A   A   S   D   V   L   F   K   E   H   E   R   I   S    443

1715 ATG TTC CAG GAT ACT ACT CTT TAC CTT GTT GAC GGG TTG ACC AAC TCC TTA GAA GGT GCC  1774
 444 M   F   Q   D   T   T   L   Y   L   V   D   G   L   T   N   S   L   E   G   A    463
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FIGURE 12-1

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1775 AGA CAA GGA ACC GCT AAA CTG ACA ACC TGG TTA GGC AAG CAG CTC GGG ATA CTA GGA AAA 1834

464 R Q G T A K L T T W L G K Q L G I L G K 483

1835 AAG TTG GAA AAC AAG AGT AAG ACG TGG TTT GGA GCA TAC GCT GCT TCC CCT TAC TGT GAT 1894

484 K L E N K S K T W F G A Y A A S P Y C D 503

1895 GTC GAT CGC AAA ATT GGC TAC ATA TGG TAT ACA AAA AAT TGC ACC CCT GCC TGC TTA CCC 1954

504 V D R K I G Y I W Y T K N C T P A C L P 523

1955 AAG AAC ACA AAA ATT GTC GGC CCT GGG AAA TTT GAC ACC AAT GCA GAG GAC GGC AAG ATA 2014

524 K N T K I V G P G K F D T N A E D G K I 543

2015 TTA CAT GAG ATG GGG GGT CAC TTG TCG GAG GTA CTA CTA CTT TCT TTA GTG GTG CTG TCC 2074

544 L H E M G G H L S E V L L L S L V V L S 563

2075 GAC TTC GCA CCG GAA ACA GCT AGT GTA ATG TAC CTA ATC CTA CAT TTT TCC ATC CCA CAA 2134

564 D F A P E T A S V M Y L I L H F S I P Q 583

2135 AGT CAC GTT GAT GTA ATG GAT TGT GAT AAG ACC CAG TTG AAC CTC ACA GTG GAG CTG ACA 2194

584 S H V D V M D C D K T Q L N L T V E L T 603

2195 ACA GCT GAA GTA ATA CCA GGG TCG GTC TGG AAT CTA GGC AAA TAT GTA TGT ATA AGA CCA 2254

604 T A E V I P G S V W N L G K Y V C I R P 623

2255 AAT TGG TGG CCT TAT GAG ACA ACT GTA GTG TTG GCA TTT GAA GAG GTG AGC CAG GTG GTG 2314

624 N W W P Y E T T V V L A F E E V S Q V V 643

2315 AAG TTA GTG TTG AGG GCA CTC AGA GAT TTA ACA CGC ATT TGG AAC GCT GCA ACA ACT ACT 2374

644 K L V L R A L R D L T R I W N A A T T T 663

2375 GCT TTT TTA GTA TGC CTT GTT AAG ATA GTC AGG GGC CAG ATG GTA CAG GGC ATT CTG TGG 2434

664 A F L V C L V K I V R G Q M V Q G I L W 683

2435 CTA CTA TTG ATA ACA GGG GTA CAA GGG CAC TTG GAT TGC AAA CCT GAA TTC TCG TAT GCC 2494

684 L L L I T G V Q G H L D C K P E F S Y A 703

2495 ATA GCA AAG GAC GAA AGA ATT GGT CAA CTG GGG GCT GAA GGC CTT ACC ACC ACT TGG AAG 2554

704 I A K D E R I G Q L G A E G L T T T W K 723

2555 GAA TAC TCA CCT GGA ATG AAG CTG GAA GAC ACA ATG GTC ATT GCT TGG TGC GAA GAT GGG 2614

724 E Y S P G M K L E D T M V I A W C E D G 743

2615 AAG TTA ATG TAC CTC CAA AGA TGC ACG AGA GAA ACC AGG TAT CTC GCA ATC TTG CAT ACA 2674

744 K L M Y L Q R C T R E T R Y L A I L H T 763

2675 AGA GCC TTG CCG ACC AGT GTG GTA TTC AAA AAA CTC TTT GAT GGG CGA AAG CAA GAG GAT 2734

764 R A L P T S V V F K K L F D G R K Q E D 783

2735 GTA GTC GAA ATG AAC GAC AAC TTT GAA TTT GGA CTC TGC CCA TGT GAT GCC AAA CCC ATA 2794

784 V V E M N D N F E F G L C P C D A K P I 803

2795 GTA AGA GGG AAG TTC AAT ACA ACG CTG CTG AAC GGA CCG GCC TTC CAG ATG GTA TGC CCC 2854

804 V R G K F N T T L L N G P A F Q M V C P 823

2855 ATA GGA TGG ACA GGG ACT GTA AGC TGT ACG TCA TTC AAT ATG GAC ACC TTA GCC ACA ACT 2914

824 I G W T G T V S C T S F N M D T L A T T 843

2915 GTG GTA CGG ACA TAT AGA AGG TCT AAA CCA TTC CCT CAT AGG CAA GGC TGT ATC ACC CAA 2974

844 V V R T Y R R S K P F P H R Q G C I T Q 863

2975 AAG AAT CTG GGG GAG GAT CTC CAT AAC TGC ATC CTT GGA GGA AAT TGG ACT TGT GTG CCT 3034

864 K N L G E D L H N C I L G G N W T C V P 883

3035 GGA GAC CAA CTA CTA TAC AAA GGG GGC TCT ATT GAA TCT TGC AAG TGG TGT GGC TAT CAA 3094

884 G D Q L L Y K G G S I E S C K W C G Y Q 903

3095 TTT AAA GAG AGT GAG GGA CTA CCA CAC TAC CCC ATT GGC AAG TGT AAA TTG GAG AAC GAG 3154

904 F K E S E G L P H Y P I G K C K L E N E 923

3155 ACT GGT TAC AGG CTA GTA GAC AGT ACC TCT TGC AAT AGA GAA GGT GTG GCC ATA GTA CCA 3214

924 T G Y R L V D S T S C N R E G V A I V P 943

3215 CAA GGG ACA TTA AAG TGC AAG ATA GGA AAA ACA ACT GTA CAG GTC ATA GCT ATG GAT ACC 3274

944 Q G T L K C K I G K T T V Q V I A M D T 963

3275 AAA CTC GGA CCT ATG CCT TGC AGA CCA TAT GAA ATC ATA TCA AGT GAG GGG CCT GTA GAA 3334

964 K L G P M P C R P Y E I I S S E G P V E 983

3335 AAG ACA GCG TGT ACT TTC AAC TAC ACT AAG ACA TTA AAA AAT AAG TAT TTT GAG CCC AGA 3394

984 K T A C T F N Y T K T L K N K Y F E P R 1003

3395 GAC AGC TAC TTT CAG CAA TAC ATG CTA AAA GGA GAG TAT CAA TAC TGG TTT GAC CTG GAG 3454

1004 D S Y F Q Q Y M L K G E Y Q Y W F D L E 1023

3455 GTG ACT GAC CAT CAC CGG GAT TAC TTC GCT GAG TCC ATA TTA GTG GTG GTA GTA GCC CTC 3514

1024 V T D H H R D Y F A E S I L V V V V A L 1043

FIGURE 12-2

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3515 TTG GGT GGC AGA TAT GTA CTT TGG TTA CTG GTT ACA TAC ATG GTC TTA TCA GAA CAG AAG 3574
1044 L   G   G   R   Y   V   L   W   L   L   V   T   Y   M   V   L   S   E   Q   K   1063

3575 GCC TTA GGG ATT CAG TAT GGA TCA GGG GAA GTG GTG ATG ATG GGC AAC TTG CTA ACC CAT 3634
1064 A   L   G   I   Q   Y   G   S   G   E   V   V   M   M   G   N   L   L   T   H   1083

3635 AAC AAT ATT GAA GTG GTG ACA TAC TTC TTG CTG CTG TAC CTA CTG CTG AGG GAG GAG AGC 3694
1084 N   N   I   E   V   V   T   Y   F   L   L   L   Y   L   L   L   R   E   E   S   1103

3695 GTA AAG AAG TGG GTC TTA CTC TTA TAC CAC ATC TTA GTG GTA CAC CCA ATC AAA TCT GTA 3754
1104 V   K   K   W   V   L   L   L   Y   H   I   L   V   V   H   P   I   K   S   V   1123

3755 ATT GTG ATC CTA CTG ATG ATT GGG GAT GTG GTA AAG GCC GAT TCA GGG GGC CAA GAG TAC 3814
1124 I   V   I   L   L   M   I   G   D   V   V   K   A   D   S   G   G   Q   E   Y   1143

3815 TTG GGG AAA ATA GAC CTC TGT TTT ACA ACA GTA GTA CTA ATC GTC ATA GGT TTA ATC ATA 3874
1144 L   G   K   I   D   L   C   F   T   T   V   V   L   I   V   I   G   L   I   I   1163

3875 GCC AGG CGT GAC CCA ACT ATA GTG CCA CTG GTA ACA ATA ATG GCA GCA CTG AGG GTC ACT 3934
1164 A   R   R   D   P   T   I   V   P   L   V   T   I   M   A   A   L   R   V   T   1183

3935 GAA CTG ACC CAC CAG CCT GGA GTT GAC ATC GCT GTG GCG GTC ATG ACT ATA ACC CTA CTG 3994
1184 E   L   T   H   Q   P   G   V   D   I   A   V   A   V   M   T   I   T   L   L   1203

3995 ATG GTT AGC TAT GTG ACA GAT TAT TTT AGA TAT AAA AAA TGG TTA CAG TGC ATT CTC AGC 4054
1204 M   V   S   Y   V   T   D   Y   F   R   Y   K   K   W   L   Q   C   I   L   S   1223

4055 CTG GTA TCT GCG GTG TTC TTG ATA AGA AGC CTA ATA TAC CTA GGT AGA ATC GAG ATG CCA 4114
1224 L   V   S   A   V   F   L   I   R   S   L   I   Y   L   G   R   I   E   M   P   1243

4115 GAG GTA ACT ATC CCA AAC TGG AGA CCA CTA ACT TTA ATA CTA TTA TAT TTG ATC TCA ACA 4174
1244 E   V   T   I   P   N   W   R   P   L   T   L   I   L   L   Y   L   I   S   T   1263

4175 ACA ATT GTA ACG AGG TGG AAG GTT GAC GTG GCT GGC CTA TTG TTG CAA TGT GTG CCT ATC 4234
1264 T   I   V   T   R   W   K   V   D   V   A   G   L   L   L   Q   C   V   P   I   1283

4235 TTA TTG CTG GTC ACA ACC TTG TGG GCC GAC TTC TTA ACC CTA ATA CTG ATC CTG CCT ACC 4294
1284 L   L   L   V   T   T   L   W   A   D   F   L   T   L   I   L   I   L   P   T   1303

4295 TAT GAA TTG GTT AAA TTA TAC TAT CTG AAA ACT GTT AGG ACT GAT ATA GAA AGA AGT TGG 4354
1304 Y   E   L   V   K   L   Y   Y   L   K   T   V   R   T   D   I   E   R   S   W   1323

4355 CTA GGG GGG ATA GAC TAT ACA AGA GTT GAC TCC ATC TAC GAC GTT GAT GAG AGT GGA GAG 4414
1324 L   G   G   I   D   Y   T   R   V   D   S   I   Y   D   V   D   E   S   G   E   1343

4415 GGC GTA TAT CTT TTT CCA TCA AGG CAG AAA GCA CAG GGG AAT TTT TCT ATA CTC TTG CCC 4474
1344 G   V   Y   L   F   P   S   R   Q   K   A   Q   G   N   F   S   I   L   L   P   1363

4475 CTT ATC AAA GCA ACA CTG ATA AGT TGC GTC AGC AGT AAA TGG CAG CTA ATA TAC ATG AGT 4534
1364 L   I   K   A   T   L   I   S   C   V   S   S   K   W   Q   L   I   Y   M   S   1383

4535 TAC TTA ACT TTG GAC TTT ATG TAC TAC ATG CAC AGG AAA GTT ATA GAA GAG ATC TCA GGA 4594
1384 Y   L   T   L   D   F   M   Y   Y   M   H   R   K   V   I   E   E   I   S   G   1403

4595 GGT ACC AAC ATA ATA TCC AGG TTA GTG GCA GCA CTC ATA GAG CTG AAC TGG TCC ATG GAA 4654
1404 G   T   N   I   I   S   R   L   V   A   A   L   I   E   L   N   W   S   M   E   1423

4655 GAA GAG GAG AGC AAA GGC TTA AAG AAG TTT TAT CTA TTG TCT GGA AGG TTG AGA AAC CTA 4714
1424 E   E   E   S   K   G   L   K   K   F   Y   L   L   S   G   R   L   R   N   L   1443

4715 ATA ATA AAA CAT AAG GTA AGG AAT GAG ACC GTG GCT TCT TGG TAC GGG GAG GAG GAA GTC 4774
1444 I   I   K   H   K   V   R   N   E   T   V   A   S   W   Y   G   E   E   E   V   1463

4775 TAC GGT ATG CCA AAG ATC ATG ACT ATA ATC AAG GCC AGT ACA CTG AGT AAG AGC AGG CAC 4834
1464 Y   G   M   P   K   I   M   T   I   I   K   A   S   T   L   S   K   S   R   H   1483

4835 TGC ATA ATA TGC ACT GTA TGT GAG GGC CGA GAG TGG AAA GGT GGC ACC TGC CCA AAA TGT 4894
1484 C   I   I   C   T   V   C   E   G   R   E   W   K   G   G   T   C   P   K   C   1503

4895 GGA CGC CAT GGG AAG CCG ATA ACG TGT GGG ATG TCG CTA GCA GAT TTT GAA GAA AGA CAC 4954
1504 G   R   H   G   K   P   I   T   C   G   M   S   L   A   D   F   E   E   R   H   1523

4955 TAT AAA AGA ATC TTT ATA AGG GAA GGC AAC TTT GAG gggccc TTC AGG CAG GAA TAC AAT 5014
1524 Y   K   R   I   F   I   R   E   G   N   F   E          F   R   Q   E   Y   N   1541

5015 GGC TTT GTA CAA TAT ACC GCT AGG GGG CAA CTA TTT CTG AGA AAC TTG CCC GTA CTG GCA 5074
1542 G   F   V   Q   Y   T   A   R   G   Q   L   F   L   R   N   L   P   V   L   A   1561

5075 ACT AAA GTA AAA ATG CTC ATG GTA GGC AAC CTT GGA GAA GAA ATT GGT AAT CTG GAA CAT 5134
1562 T   K   V   K   M   L   M   V   G   N   L   G   E   E   I   G   N   L   E   H   1581

5135 CTT GGG TGG ATC CTA AGG GGG CCT GCC GTG TGT AAG AAG ATC ACA GAG CAC GAA AAA TGC 5194
1582 L   G   W   I   L   R   G   P   A   V   C   K   K   I   T   E   H   E   K   C   1601

5195 CAC ATT AAT ATA CTG GAT AAA CTA ACC GCA TTT TTC GGG ATC ATG CCA AGG GGG ACT ACA 5254
1602 H   I   N   I   L   D   K   L   T   A   F   F   G   I   M   P   R   G   T   T   1621
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FIGURE 12-3

BVDV NADL clns- (inf. clone) Genes 4/21/99 5:45:24 PM Page 4

5255 CCC AGA GCC CCG GTG AGG TTC CCT ACG AGC TTA CTA AAA GTG AGG AGG GGT CTG GAG ACT 5314
1622 P R A P V R F P T S L L K V R R G L E T 1641

5315 GCC TGG GCT TAC ACA CAC CAA GGC GGG ATA AGT TCA GTC GAC CAT GTA ACC GCC GGA AAA 5374
1642 A W A Y T H Q G G I S S V D H V T A G K 1661

5375 GAT CTA CTG GTC TGT GAC AGC ATG GGA CGA ACT AGA GTG GTT TGC CAA AGC AAC AAC AGG 5434
1662 D L L V C D S M G R T R V V C Q S N N R 1681

5435 TTG ACC GAT GAG ACA GAG TAT GGC GTC AAG ACT GAC TCA GGG TGC CCA GAC GGT GCC AGA 5494
1682 L T D E T E Y G V K T D S G C P D G A R 1701

5495 TGT TAT GTG TTA AAT CCA GAG GCC GTT AAC ATA TCA GGA TCC AAA GGG GCA GTC GTT CAC 5554
1702 C Y V L N P E A V N I S G S K G A V V H 1721

5555 CTC CAA AAG ACA GGT GGA GAA TTC ACG TGT GTC ACC GCA TCA GGC ACA CCG GCT TTC TTC 5614
1722 L Q K T G G E F T C V T A S G T P A F F 1741

5615 GAC CTA AAA AAC TTG AAA GGA TGG TCA GGC TTG CCT ATA TTT GAA GCC TCC AGC GGG AGG 5674
1742 D L K N L K G W S G L P I F E A S S G R 1761

5675 GTG GTT GGC AGA GTC AAA GTA GGG AAG AAT GAA GAG TCT AAA CCT ACA AAA ATA ATG AGT 5734
1762 V V G R V K V G K N E E S K P T K I M S 1781

5735 GGA ATC CAG ACC GTC TCA AAA AAC AGA GCA GAC CTG ACC GAG ATG GTC AAG AAG ATA ACC 5794
1782 G I Q T V S K N R A D L T E M V K K I T 1801

5795 AGC ATG AAC AGG GGA GAC TTC AAG CAG ATT ACT TTG GCA ACA GGG GCA GGC AAA ACC ACA 5854
1802 S M N R G D F K Q I T L A T G A G K T T 1821

5855 GAA CTC CCA AAA GCA GTT ATA GAG GAG ATA GGA AGA CAC AAG AGA GTA TTA GTT CTT ATA 5914
1822 E L P K A V I E E I G R H K R V L V L I 1841

5915 CCA TTA AGG GCA GCG GCA GAG TCA GTC TAC CAG TAT ATG AGA TTG AAA CAC CCA AGC ATC 5974
1842 P L R A A A E S V Y Q Y M R L K H P S I 1861

5975 TCT TTT AAC CTA AGG ATA GGG GAC ATG AAA GAG GGG GAC ATG GCA ACC GGG ATA ACC TAT 6034
1862 S F N L R I G D M K E G D M A T G I T Y 1881

6035 GCA TCA TAC GGG TAC TTC TGC CAA ATG CCT CAA CCA AAG CTC AGA GCT GCT ATG GTA GAA 6094
1882 A S Y G Y F C Q M P Q P K L R A A M V E 1901

6095 TAC TCA TAC ATA TTC TTA GAT GAA TAC CAT TGT GCC ACT CCT GAA CAA CTG GCA ATT ATC 6154
1902 Y S Y I F L D E Y H C A T P E Q L A I I 1921

6155 GGG AAG ATC CAC AGA TTT TCA GAG AGT ATA AGG GTT GTC GCC ATG ACT GCC ACG CCA GCA 6214
1922 G K I H R F S E S I R V V A M T A T P A 1941

6215 GGG TCG GTG ACC ACA ACA GGT CAA AAG CAC CCA ATA GAG GAA TTC ATA GCC CCC GAG GTA 6274
1942 G S V T T T G Q K H P I E E F I A P E V 1961

6275 ATG AAA GGG GAG GAT CTT GGT AGT CAG TTC CTT GAT ATA GCA GGG TTA AAA ATA CCA GTG 6334
1962 M K G E D L G S Q F L D I A G L K I P V 1981

6335 GAT GAG ATG AAA GGC AAT ATG TTG GTT TTT GTA CCA ACG AGA AAC ATG GCA GTA GAG GTA 6394
1982 D E M K G N M L V F V P T R N M A V E V 2001

6395 GCA AAG AAG CTA AAA GCT AAG GGC TAT AAC TCT GGA TAC TAT TAC AGT GGA GAG GAT CCA 6454
2002 A K K L K A K G Y N S G Y Y Y S G E D P 2021

6455 GCC AAT CTG AGA GTT GTG ACA TCA CAA TCC CCC TAT GTA ATC GTG GCT ACA AAT GCT ATT 6514
2022 A N L R V V T S Q S P Y V I V A T N A I 2041

6515 GAA TCA GGA GTG ACA CTA CCA GAT TTG GAC ACG GTT ATA GAC ACG GGG TTG AAA TGT GAA 6574
2042 E S G V T L P D L D T V I D T G L K C E 2061

6575 AAG AGG GTG AGG GTA TCA TCA AAG ATA CCC TTC ATC GTA ACA GGC CTT AAG AGG ATG GCC 6634
2062 K R V R V S S K I P F I V T G L K R M A 2081

6635 GTG ACT GTG GGT GAG CAG GCG CAG CGT AGG GGC AGA GTA GGT AGA GTG AAA CCC GGG AGG 6694
2082 V T V G E Q A Q R R G R V G R V K P G R 2101

6695 TAT TAT AGG AGC CAG GAA ACA GCA ACA GGG TCA AAG GAC TAC CAC TAT GAC CTC TTG CAG 6754
2102 Y Y R S Q E T A T G S K D Y H Y D L L Q 2121

6755 GCA CAA AGA TAC GGG ATT GAG GAT GGA ATC AAC GTG ACG AAA TCC TTT AGG GAG ATG AAT 6814
2122 A Q R Y G I E D G I N V T K S F R E M N 2141

6815 TAC GAT TGG AGC CTA TAC GAG GAG GAC AGC CTA CTA ATA ACC CAG CTG GAA ATA CTA AAT 6874
2142 Y D W S L Y E E D S L L I T Q L E I L N 2161

6875 AAT CTA CTC ATC TCA GAA GAC TTG CCA GCC GCT GTT AAG AAC ATA ATG GCC AGG ACT GAT 6934
2162 N L L I S E D L P A A V K N I M A R T D 2181

6935 CAC CCA GAG CCA ATC CAA CTT GCA TAC AAC AGC TAT GAA GTC CAG GTC CCG GTC CTG TTC 6994
2182 H P E P I Q L A Y N S Y E V Q V P V L F 2201

FIGURE 12-4

6995 CCA AAA ATA AGG AAT GGA GAA GTC ACA GAC ACC TAC GAA AAT TAC TCG TTT CTA AAT GCC 7054
2202 P K I R N G E V T D T Y E N Y S F L N A 2221

7055 AGA AAG TTA GGG GAG GAT GTG CCC GTG TAT ATC TAC GCT ACT GAA GAT GAG GAT CTG GCA 7114
2222 R K L G E D V P V Y I Y A T E D E D L A 2241

7115 GTT GAC CTC TTA GGG CTA GAC TGG CCT GAT CCT GGG AAC CAG CAG GTA GTG GAG ACT GGT 7174
2242 V D L L G L D W P D P G N Q Q V V E T G 2261

7175 AAA GCA CTG AAG CAA GTG ACC GGG TTG TCC TCG GCT GAA AAT GCC CTA CTA GTG GCT TTA 7234
2262 K A L K Q V T G L S S A E N A L L V A L 2281

7235 TTT GGG TAT GTG GGT TAC CAG GCT CTC TCA AAG AGG CAT GTC CCA ATG ATA ACA GAC ATA 7294
2282 F G Y V G Y Q A L S K R H V P M I T D I 2301

7295 TAT ACC ATC GAG GAC CAG AGA CTA GAA GAC ACC ACC CAC CTC CAG TAT GCA CCC AAC GCC 7354
2302 Y T I E D Q R L E D T T H L Q Y A P N A 2321

7355 ATA AAA ACC GAT GGG ACA GAG ACT GAA CTG AAA GAA CTG GCG TCG GGT GAC GTG GAA AAA 7414
2322 I K T D G T E T E L K E L A S G D V E K 2341

7415 ATC ATG GGA GCC ATT TCA GAT TAT GCA GCT GGG GGA CTG GAG TTT GTT AAA TCC CAA GCA 7474
2342 I M G A I S D Y A A G G L E F V K S Q A 2361

7475 GAA AAG ATA AAA ACA GCT CCT TTG TTT AAA GAA AAC GCA GAA GCC GCA AAA GGG TAT GTC 7534
2362 E K I K T A P L F K E N A E A A K G Y V 2381

7535 CAA AAA TTC ATT GAC TCA TTA ATT GAA AAT AAA GAA GAA ATA ATC AGA TAT GGT TTG TGG 7594
2382 Q K F I D S L I E N K E E I I R Y G L W 2401

7595 GGA ACA CAC ACA GCA CTA TAC AAA AGC ATA GCT GCA AGA CTG GGG CAT GAA ACA GCG TTT 7654
2402 G T H T A L Y K S I A A R L G H E T A F 2421

7655 GCC ACA CTA GTG TTA AAG TGG CTA GCT TTT GGA GGG GAA TCA GTG TCA GAC CAC GTC AAG 7714
2422 A T L V L K W L A F G G E S V S D H V K 2441

7715 CAG GCG GCA GTT GAT TTA GTG GTC TAT TAT GTG ATG AAT AAG CCT TCC TTC CCA GGT GAC 7774
2442 Q A A V D L V V Y Y V M N K P S F P G D 2461

7775 TCC GAG ACA CAG CAA GAA GGG AGG CGA TTC GTC GCA AGC CTG TTC ATC TCC GCA CTG GCA 7834
2462 S E T Q Q E G R R F V A S L F I S A L A 2481

7835 ACC TAC ACA TAC AAA ACT TGG AAT TAC CAC AAT CTC TCT AAA GTG GTG GAA CCA GCC CTG 7894
2482 T Y T Y K T W N Y H N L S K V V E P A L 2501

7895 GCT TAC CTC CCC TAT GCT ACC AGC GCA TTA AAA ATG TTC ACC CCA ACG CGG CTG GAG AGC 7954
2502 A Y L P Y A T S A L K M F T P T R L E S 2521

7955 GTG GTG ATA CTG AGC ACC ACG ATA TAT AAA ACA TAC CTC TCT ATA AGG AAG GGG AAG AGT 8014
2522 V V I L S T T I Y K T Y L S I R K G K S 2541

8015 GAT GGA TTG CTG GGT ACG GGG ATA AGT GCA GCC ATG GAA ATC CTG TCA CAA AAC CCA GTA 8074
2542 D G L L G T G I S A A M E I L S Q N P V 2561

8075 TCG GTA GGT ATA TCT GTG ATG TTG GGG GTA GGG GCA ATC GCT GCG CAC AAC GCT ATT GAG 8134
2562 S V G I S V M L G V G A I A A H N A I E 2581

8135 TCC AGT GAA CAG AAA AGG ACC CTA CTT ATG AAG GTG TTT GTA AAG AAC TTC TTG GAT CAG 8194
2582 S S E Q K R T L L M K V F V K N F L D Q 2601

8195 GCT GCA ACA GAT GAG CTG GTA AAA GAA AAC CCA GAA AAA ATT ATA ATG GCC TTA TTT GAA 8254
2602 A A T D E L V K E N P E K I I M A L F E 2621

8255 GCA GTC CAG ACA ATT GGT AAC CCC CTG AGA CTA ATA TAC CAC CTG TAT GGG GTT TAC TAC 8314
2622 A V Q T I G N P L R L I Y H L Y G V Y Y 2641

8315 AAA GGT TGG GAG GCC AAG GAA CTA TCT GAG AGG ACA GCA GGC AGA AAC TTA TTC ACA TTG 8374
2642 K G W E A K E L S E R T A G R N L F T L 2661

8375 ATA ATG TTT GAA GCC TTC GAG TTA TTA GGG ATG GAC TCA CAA GGG AAA ATA AGG AAC CTG 8434
2662 I M F E A F E L L G M D S Q G K I R N L 2681

8435 TCC GGA AAT TAC ATT TTG GAT TTG ATA TAC GGC CTA CAC AAG CAA ATC AAC AGA GGG CTG 8494
2682 S G N Y I L D L I Y G L H K Q I N R G L 2701

8495 AAG AAA ATG GTA CTG GGG TGG GCC CCT GCA CCC TTT AGT TGT GAC TGG ACC CCT AGT GAC 8554
2702 K K M V L G W A P A P F S C D W T P S D 2721

8555 GAG AGG ATC AGA TTG CCA ACA GAC AAC TAT TTG AGG GTA GAA ACC AGG TGC CCA TGT GGC 8614
2722 E R I R L P T D N Y L R V E T R C P C G 2741

8615 TAT GAG ATG AAA GCT TTC AAA AAT GTA GGT GGC AAA CTT ACC AAA GTG GAG GAG AGC GGG 8674
2742 Y E M K A F K N V G G K L T K V E E S G 2761

8675 CCT TTC CTA TGT AGA AAC AGA CCT GGT AGG GGA CCA GTC AAC TAC AGA GTC ACC AAG TAT 8734
2762 P F L C R N R P G R G P V N Y R V T K Y 2781

FIGURE 12-5

8735 TAC GAT GAC AAC CTC AGA GAG ATA AAA CCA GTA GCA AAG TTG GAA GGA CAG GTA GAG CAC 8794
2782 Y D D N L R E I K P V A K L E G Q V E H 2801

8795 TAC TAC AAA GGG GTC ACA GCA AAA ATT GAC TAC AGT AAA GGA AAA ATG CTC TTG GCC ACT 8854
2802 Y Y K G V T A K I D Y S K G K M L L A T 2821

8855 GAC AAG TGG GAG GTG GAA CAT GGT GTC ATA ACC AGG TTA GCT AAG AGA TAT ACT GGG GTC 8914
2822 D K W E V E H G V I T R L A K R Y T G V 2841

8915 GGG TTC AAT GGT GCA TAC TTA GGT GAC GAG CCC AAT CAC CGT GCT CTA GTG GAG AGG GAC 8974
2842 G F N G A Y L G D E P N H R A L V E R D 2861

8975 TGT GCA ACT ATA ACC AAA AAC ACA GTA CAG TTT CTA AAA ATG AAG AAG GGG TGT GCG TTC 9034
2862 C A T I T K N T V Q F L K M K K G C A F 2881

9035 ACC TAT GAC CTG ACC ATC TCC AAT CTG ACC AGG CTC ATC GAA CTA GTA CAC AGG AAC AAT 9094
2882 T Y D L T I S N L T R L I E L V H R N N 2901

9095 CTT GAA GAG AAG GAA ATA CCC ACC GCT ACG GTC ACC ACA TGG CTA GCT TAC ACC TTC GTG 9154
2902 L E E K E I P T A T V T T W L A Y T F V 2921

9155 AAT GAA GAC GTA GGG ACT ATA AAA CCA GTA CTA GGA GAG AGA GTA ATC CCC GAC CCT GTA 9214
2922 N E D V G T I K P V L G E R V I P D P V 2941

9215 GTT GAT ATC AAT TTA CAA CCA GAG GTG CAA GTG GAC ACG TCA GAG GTT GGG ATC ACA ATA 9274
2942 V D I N L Q P E V Q V D T S E V G I T I 2961

9275 ATT GGA AGG GAA ACC CTG ATG ACA ACG GGA GTG ACA CCT GTC TTG GAA AAA GTA GAG CCT 9334
2962 I G R E T L M T T G V T P V L E K V E P 2981

9335 GAC GCC AGC GAC AAC CAA AAC TCG GTG AAG ATC GGG TTG GAT GAG GGT AAT TAC CCA GGG 9394
2982 D A S D N Q N S V K I G L D E G N Y P G 3001

9395 CCT GGA ATA CAG ACA CAT ACA CTA ACA GAA GAA ATA CAC AAC AGG GAT GCG AGG CCC TTC 9454
3002 P G I Q T H T L T E E I H N R D A R P F 3021

9455 ATC ATG ATC CTG GGC TCA AGG AAT TCC ATA TCA AAT AGG GCA AAG ACT GCT AGA AAT ATA 9514
3022 I M I L G S R N S I S N R A K T A R N I 3041

9515 AAT CTG TAC ACA GGA AAT GAC CCC AGG GAA ATA CGA GAC TTG ATG GCT GCA GGG CGC ATG 9574
3042 N L Y T G N D P R E I R D L M A A G R M 3061

9575 TTA GTA GTA GCA CTG AGG GAT GTC GAC CCT GAG CTG TCT GAA ATG GTC GAT TTC AAG GGG 9634
3062 L V V A L R D V D P E L S E M V D F K G 3081

9635 ACT TTT TTA GAT AGG GAG GCC CTG GAG GCT CTA AGT CTC GGG CAA CCT AAA CCG AAG CAG 9694
3082 T F L D R E A L E A L S L G Q P K P K Q 3101

9695 GTT ACC AAG GAA GCT GTT AGG AAT TTG ATA GAA CAG AAA AAA GAT GTG GAG ATC CCT AAC 9754
3102 V T K E A V R N L I E Q K K D V E I P N 3121

9755 TGG TTT GCA TCA GAT GAC CCA GTA TTT CTG GAA GTG GCC TTA AAA AAT GAT AAG TAC TAC 9814
3122 W F A S D D P V F L E V A L K N D K Y Y 3141

9815 TTA GTA GGA GAT GTT GGA GAG CTA AAA GAT CAA GCT AAA GCA CTT GGG GCC ACG GAT CAG 9874
3142 L V G D V G E L K D Q A K A L G A T D Q 3161

9875 ACA AGA ATT ATA AAG GAG GTA GGC TCA AGG ACG TAT GCC ATG AAG CTA TCT AGC TGG TTC 9934
3162 T R I I K E V G S R T Y A M K L S S W F 3181

9935 CTC AAG GCA TCA AAC AAA CAG ATG AGT TTA ACT CCA CTG TTT GAG GAA TTG TTG CTA CGG 9994
3182 L K A S N K Q M S L T P L F E E L L L R 3201

9995 TGC CCA CCT GCA ACT AAG AGC AAT AAG GGG CAC ATG GCA TCA GCT TAC CAA TTG GCA CAG 10054
3202 C P P A T K S N K G H M A S A Y Q L A Q 3221

10055 GGT AAC TGG GAG CCC CTC GGT TGC GGG GTG CAC CTA GGT ACA ATA CCA GCC AGA AGG GTG 10114
3222 G N W E P L G C G V H L G T I P A R R V 3241

10115 AAG ATA CAC CCA TAT GAA GCT TAC CTG AAG TTG AAA GAT TTC ATA GAA GAA GAA GAG AAG 10174
3242 K I H P Y E A Y L K L K D F I E E E E K 3261

10175 AAA CCT AGG GTT AAG GAT ACA GTA ATA AGA GAG CAC AAC AAA TGG ATA CTT AAA AAA ATA 10234
3262 K P R V K D T V I R E H N K W I L K K I 3281

10235 AGG TTT CAA GGA AAC CTC AAC ACC AAG AAA ATG CTC AAC CCG GGG AAA CTA TCT GAA CAG 10294
3282 R F Q G N L N T K K M L N P G K L S E Q 3301

10295 TTG GAC AGG GAG GGG CGC AAG AGG AAC ATC TAC AAC CAC CAG ATT GGT ACT ATA ATG TCA 10354
3302 L D R E G R K R N I Y N H Q I G T I M S 3321

10355 AGT GCA GGC ATA AGG CTG GAG AAA TTG CCA ATA GTG AGG GCC CAA ACC GAC ACC AAA ACC 10414
3322 S A G I R L E K L P I V R A Q T D T K T 3341

10415 TTT CAT GAG GCA ATA AGA GAT AAG ATA GAC AAG AGT GAA AAC CGG CAA AAT CCA GAA TTG 10474
3342 F H E A I R D K I D K S E N R Q N P E L 3361

FIGURE 12-6

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10475 CAC AAC AAA TTG TTG GAG ATT TTC CAC ACG ATA GCC CAA CCC ACC CTG AAA CAC ACC TAC 10534
 3362 H   N   K   L   L   E   I   F   H   T   I   A   Q   P   T   L   K   H   T   Y   3381

10535 GGT GAG GTG ACG TGG GAG CAA CTT GAG GCG GGG ATA AAT AGA AAG GGG GCA GCA GGC TTC 10594
 3382 G   E   V   T   W   E   Q   L   E   A   G   I   N   R   K   G   A   A   G   F   3401

10595 CTG GAG AAG AAG AAC ATC GGA GAA GTA TTG GAT TCA GAA AAG CAC CTG GTA GAA CAA TTG 10654
 3402 L   E   K   K   N   I   G   E   V   L   D   S   E   K   H   L   V   E   Q   L   3421

10655 GTC AGG GAT CTG AAG GCC GGG AGA AAG ATA AAA TAT TAT GAA ACT GCA ATA CCA AAA AAT 10714
 3422 V   R   D   L   K   A   G   R   K   I   K   Y   Y   E   T   A   I   P   K   N   3441

10715 GAG AAG AGA GAT GTC AGT GAT GAC TGG CAG GCA GGG GAC CTG GTG GTT GAG AAG AGG CCA 10774
 3442 E   K   R   D   V   S   D   D   W   Q   A   G   D   L   V   V   E   K   R   P   3461

10775 AGA GTT ATC CAA TAC CCT GAA GCC AAG ACA AGG CTA GCC ATC ACT AAG GTC ATG TAT AAC 10834
 3462 R   V   I   Q   Y   P   E   A   K   T   R   L   A   I   T   K   V   M   Y   N   3481

10835 TGG GTG AAA CAG CAG CCC GTT GTG ATT CCA GGA TAT GAA GGA AAG ACC CCC TTG TTC AAC 10894
 3482 W   V   K   Q   Q   P   V   V   I   P   G   Y   E   G   K   T   P   L   F   N   3501

10895 ATC TTT GAT AAA GTG AGA AAG GAA TGG GAC TCG TTC AAT GAG CCA GTG GCC GTA AGT TTT 10954
 3502 I   F   D   K   V   R   K   E   W   D   S   F   N   E   P   V   A   V   S   F   3521

10955 GAC ACC AAA GCC TGG GAC ACT CAA GTG ACT AGT AAG GAT CTG CAA CTT ATT GGA GAA ATC 11014
 3522 D   T   K   A   W   D   T   Q   V   T   S   K   D   L   Q   L   I   G   E   I   3541

11015 CAG AAA TAT TAC TAT AAG AAG GAG TGG CAC AAG TTC ATT GAC ACC ATC ACC GAC CAC ATG 11074
 3542 Q   K   Y   Y   Y   K   K   E   W   H   K   F   I   D   T   I   T   D   H   M   3561

11075 ACA GAA GTA CCA GTT ATA ACA GCA GAT GGT GAA GTA TAT ATA AGA AAT GGG CAG AGA GGG 11134
 3562 T   E   V   P   V   I   T   A   D   G   E   V   Y   I   R   N   G   Q   R   G   3581

11135 AGC GGC CAG CCA GAC ACA AGT GCT GGC AAC AGC ATG TTA AAT GTC CTG ACA ATG ATG TAC 11194
 3582 S   G   Q   P   D   T   S   A   G   N   S   M   L   N   V   L   T   M   M   Y   3601

11195 GGC TTC TGC GAA AGC ACA GGG GTA CCG TAC AAG AGT TTC AAC AGG GTG GCA AGG ATC CAC 11254
 3602 G   F   C   E   S   T   G   V   P   Y   K   S   F   N   R   V   A   R   I   H   3621

11255 GTC TGT GGG GAT GAT GGC TTC TTA ATA ACT GAA AAA GGG TTA GGG CTG AAA TTT GCT AAC 11314
 3622 V   C   G   D   D   G   F   L   I   T   E   K   G   L   G   L   K   F   A   N   3641

11315 AAA GGG ATG CAG ATT CTT CAT GAA GCA GGC AAA CCT CAG AAG ATA ACG GAA GGG GAA AAG 11374
 3642 K   G   M   Q   I   L   H   E   A   G   K   P   Q   K   I   T   E   G   E   K   3661

11375 ATG AAA GTT GCC TAT AGA TTT GAG GAT ATA GAG TTC TGT TCT CAT ACC CCA GTC CCT GTT 11434
 3662 M   K   V   A   Y   R   F   E   D   I   E   F   C   S   H   T   P   V   P   V   3681

11435 AGG TGG TCC GAC AAC ACC AGT AGT CAC ATG GCC GGG AGA GAC ACC GCT GTG ATA CTA TCA 11494
 3682 R   W   S   D   N   T   S   S   H   M   A   G   R   D   T   A   V   I   L   S   3701

11495 AAG ATG GCA ACA AGA TTG GAT TCA AGT GGA GAG AGG GGT ACC ACA GCA TAT GAA AAA GCG 11554
 3702 K   M   A   T   R   L   D   S   S   G   E   R   G   T   T   A   Y   E   K   A   3721

11555 GTA GCC TTC AGT TTC TTG CTG ATG TAT TCC TGG AAC CCG CTT GTT AGG AGG ATT TGC CTG 11614
 3722 V   A   F   S   F   L   L   M   Y   S   W   N   P   L   V   R   R   I   C   L   3741

11615 TTG GTC CTT TCG CAA CAG CCA GAG ACA GAC CCA TCA AAA CAT GCC ACT TAT TAT TAC AAA 11674
 3742 L   V   L   S   Q   Q   P   E   T   D   P   S   K   H   A   T   Y   Y   Y   K   3761

11675 GGT GAT CCA ATA GGG GCC TAT AAA GAT GTA ATA GGT CGG AAT CTA AGT GAA CTG AAG AGA 11734
 3762 G   D   P   I   G   A   Y   K   D   V   I   G   R   N   L   S   E   L   K   R   3781

11735 ACA GGC TTT GAG AAA TTG GCA AAT CTA AAC CTA AGC CTG TCC ACG TTG GGG ATC TGG ACT 11794
 3782 T   G   F   E   K   L   A   N   L   N   L   S   L   S   T   L   G   I   W   T   3801

11795 AAG CAC ACA AGC AAA AGA ATA ATT CAG GAC TGT GTT GCC ATT GGG AAA GAA GAG GGC AAC 11854
 3802 K   H   T   S   K   R   I   I   Q   D   C   V   A   I   G   K   E   E   G   N   3821

11855 TGG CTA GTT AAC GCC GAC AGG CTG ATA TCC AGC AAA ACT GGC CAC TTA TAC ATA CCT GAT 11914
 3822 W   L   V   N   A   D   R   L   I   S   S   K   T   G   H   L   Y   I   P   D   3841

11915 AAA GGC TTT ACA TTA CAA GGA AAG CAT TAT GAG CAA CTG CAG CTA AGA ACA GAG ACA AAC 11974
 3842 K   G   F   T   L   Q   G   K   H   Y   E   Q   L   Q   L   R   T   E   T   N   3861

11975 CCG GTC ATG GGG GTT GGG ACT GAG AGA TAC AAG TTA GGT CCC ATA GTC AAT CTG CTG CTG 12034
 3862 P   V   M   G   V   G   T   E   R   Y   K   L   G   P   I   V   N   L   L   L   3881

12035 AGA AGG TTG AAA ATT CTG CTC ATG ACG GCC GTC GGC GTC AGC AGC TGA gacaaaatgtatatat 12098
 3882 R   R   L   K   I   L   L   M   T   A   V   G   V   S   S   *                    3897

12099 tgtaaataaattaatccatgtacatagtgtatataaatatagttgggaccgtccacctcaagaagacgacacgcccaaca 12178

12179 cgcacagctaaacagtagtcaagattatctacctcaagataacactacatttaatgcacacagcactttagctgtatgag 12258

12259 gatacgcccgacgtctatagttggactagggaagacctctaacagccccc                               12308
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FIGURE 12-7

GTATaatcactcccctgtgaggaactactgtcttcacgcagaaagcgtctagccatggcgttagtatgagtgtcgtgcagcctccag
gacccccccctcccgggagagccatagtggtctgcggaaccggtgagtacaccggaattgccaggacgaccgggtcctttcttggata
aacccgctcaatgcctggagatttgggcgtgcccccgcaagactgctagccgagtagtgttgggtcgcgaaaggccttgtggtactgc
ctgatagggtgcttgcgagtgccccgggaggtctcgtagaccgtgcaccATG

FIGURE 13

GTaatcactcccctgtgaggaactactgtcttcacgcagaaagcgtctagccatggcgttagtatgagtgtcgtgcagcctccaggac
cccccctcccgggagagccatagtggtctgcggaaccggtgagtacaccggaattgccaggacgaccgggtcctttcttggataaac
ccgctcaatgcctggagatttgggcgtgcccccgcaagactgctagccgagtagtgttgggtcgcgaaaggccttgtggtactgcctg
atagggtgcttgcgagtgccccgggaggtctcgtagaccgtgcaccATG

FIGURE 14

GTATacactccaccatgaatcactcccctgtgaggaactactgtcttcacgcagaaagcgtctagccatggcgttagtatgagtgtcg
tgcagcctccaggaccccccctcccgggagagccatagtggtctgcggaaccggtgagtacaccggaattgccaggacgaccggg
tcctttcttggataaacccgctcaatgcctggagatttgggcgtgcccccgcaagactgctagccgagtagtgttgggtcgcgaaaggc
cttgtggtactgcctgatagggtgcttgcgagtgccccgggaggtctcgtagaccgtgcaccATG

FIGURE 15

GTATCAGAAGTGCGAATGCTGAacactccaccatgaatcactcccctgtgaggaactactgtcttcacgcagaaa
gcgtctagccatggcgttagtatgagtgtcgtgcagcctccaggaccccccctcccgggagagccatagtggtctgcggaaccggtg
agtacaccggaattgccaggacgaccgggtcctttcttggataaacccgctcaatgcctggagatttgggcgtgcccccgcaagactg
ctagccgagtagtgttgggtcgcgaaaggccttgtggtactgcctgatagggtgcttgcgagtgccccgggaggtctcgtagaccgtg
caccATG

FIGURE 16

GTATgccagccccctgatgggggcgacactccaccatgaatcactcccctgtgaggaactactgtcttcacgcagaaagcgtctag
ccatggcgttagtatgagtgtcgtgcagcctccaggaccccccctcccgggagagccatagtggtctgcggaaccggtgagtacacc
ggaattgccaggacgaccgggtcctttcttggataaacccgctcaatgcctggagatttgggcgtgcccccgcaagactgctagccga
gtagtgttgggtcgcgaaaggccttgtggtactgcctgatagggtgcttgcgagtgccccgggaggtctcgtagaccgtgcaccAT
G

FIGURE 17

GTATTGCAGTTTgccagccccctgatgggggcgacactccaccatgaatcactcccctgtgaggaactactgtcttcacgc
agaaagcgtctagccatggcgttagtatgagtgtcgtgcagcctccaggaccccccctcccgggagagccatagtggtctgcggaac
cggtgagtacaccggaattgccaggacgaccgggtcctttcttggataaacccgctcaatgcctggagatttgggcgtgcccccgcaa
gactgctagccgagtagtgttgggtcgcgaaaggccttgtggtactgcctgatagggtgcttgcgagtgccccgggaggtctcgtaga
ccgtgcaccATG

FIGURE 18

GTATTGCAGTTTgccagccccctgatgggggcgacactccaccatgaatcactcccctgtgaggaactactgtcttcacgc
agaaagcgtctagccatggcgttagtatgagtgtcgtgcagcctccaggaccccccctcccgggagagccatagtggtctgcggaac
cggtgagtacaccggaattgccaggacgaccgggtcctttcttggataaacccgctcaatgcctggagatttgggcgtgcccccgcaa
gactgctagccgagtagtgttgggtcgcgaaaggccttgtggtactgcctgatagggtgcttgcgagtgccccgggaggtctcgtaga
ccgtgcaccATGGAGTTGATCACAAATGAACTTTTATACAAAACATACAAACAAAAAC
CCGTCGGGGTGGAGGAACCTGTTTATGATCAGGCAGGTGATCCCTTATTTGGT
GAAAGGGGAGCAGTCCACCCTCAATCGACGCTAAAGCTCCCACACAAGAGAG
GGGAACGCGATGTTCCAACCAACTTGGCATCCTTACCAAAAAGAGGTGACTGC
AGGTCGGGTAATAGCAGAGGACCTGTGAGCGGGATCTACCTGAAGCCAGGGC
CACTATTTTACCAGGACTATAAAGGTCCCGTCTATCACAGGGCCCCGCTGGAGC
TCTTTGAGGAGGGATCCATGTGTGAAACGACTAAACGGATAGGGAGAGTAACT
GGAAGTGACGGAAAGCTGTACCACATTTATGTGTGTATAGATGGATGTATAATA
ATAAAAAGTGCCACGAGAAGTTACCAAAGGGTGTTCAGGTGGGTCCATAATAG
GCTTGACTGCCCTCTATGGGTCACAACTTGCTCAGACACGAAAGAAGAGGGAG
CAACAAAAAAGAAAACACAGAAACCCGACAGACTAGAAAGGGGGAAAATGAA
AATAGTGCCCAAAGAATCTGAAAAAGACAGCAAAACTAAACCTCCGGATGCTA
CAATAGTGGTGGAAGGAGTCAAATACCAGGTGAGGAAGAAGGGAAAAACCAA
GAGTAAAAACACTCAGGACGGCTTGTACCATAACAAAAACAAACCTCAGGAAT
CACGCAAGAAACTGGAAAAAGCATTGTTGGCGTGGGCAATAATAGCTATAGTT
TTGTTTCAAGTTACAATGGGAGAAAACATAACACAGTGGAACCTACAAGATAAT
GGGACGGAAGGGATACAACGGGCAATGTTCCAAAGGGGTGTGAATAGAAGTT
TACATGGAATCTGGCCAGAGAAAATCTGTACTGGTGTCCCTTCCCATCTAGCCA
CCGATATAGAACTAAAAACAATTCATGGTATGATGGATGCAAGTGAGAAGACC
AACTACACGTGTTGCAGACTTCAACGCCATGAGTGGAACAAGCATGGTTGGTG
CAACTGGTACAATATTGAACCCTGGATTCTAGTCATGAATAGAACCCAAGCCAA
TCTCACTGAGGGACAACCACCAAGGGAGTGCGCAGTCACTTGTAGGTATGATA
GGGCTAGTGACTTAAACGTGGTAACACAAGCTAGAGATAGCCCCACACCCTTA
ACAGGTTGCAAGAAAGGAAAGAACTTCTCCTTTGCAGGCATATTGATGCGGGG
CCCCTGCAACTTTGAAATAGCTGCAAGTGATGTATTATTCAAAGAACATGAACG
CATTAGTATGTTCCAGGATACTACTCTTTACCTTGTTGACGGGTTGACCAACTCC
TTAGAAGGTGCCAGACAAGGAACCGCTAAACTGACAACCTGGTTAGGCAAGCA
GCTCGGGATACTAGGAAAAAAGTTGGAAAACAAGAGTAAGACGTGGTTTGGAG
CATACGCTGCTTCCCCTTACTGTGATGTCGATCGCAAAATTGGCTACATATGGT
ATACAAAAAATTGCACCCCTGCCTGCTTACCCAAGAACACAAAAATTGTCGGCC
CTGGGAAATTTGACACCAATGCAGAGGACGGCAAGATATTACATGAGATGGGG
GGTCACTTGTCGGAGGTACTACTACTTTCTTTAGTGGTGCTGTCCGACTTCGCA
CCGGAAACAGCTAGTGTAATGTACCTAATCCTACATTTTTCCATCCCACAAAGTC
ACGTTGATGTAATGGATTGTGATAAGACCCAGTTGAACCTCACAGTGGAGCTG
ACAACAGCTGAAGTAATACCAGGGTCGGTCTGGAATCTAGGCAAATATGTATG
TATAAGACCAAATTGGTGGCCTTATGAGACAACTGTAGTGTTGGCATTTGAAGA
GGTGAGCCAGGTGGTGAAGTTAGTGTTGAGGGCACTCAGAGATTTAACACGCA
TTTGGAACGCTGCAACAACTACTGCTTTTTTAGTATGCCTTGTTAAGATAGTCAG
GGGCCAGATGGTACAGGGCATTCTGTGGCTACTATTGATAACAGGGGTACAAG
GGCACTTGGATTGCAAACCTGAATTCTCGTATGCCATAGCAAAGGACGAAAGA
ATTGGTCAACTGGGGGCTGAAGGCCTTACCACCACTTGGAAGGAATACTCACC
TGGAATGAAGCTGGAAGACACAATGGTCATTGCTTGGTGCGAAGATGGGAAGT
TAATGTACCTCCAAAGATGCACGAGAGAAACCAGGTATCTCGCAATCTTGCATA
CAAGAGCCTTGCCGACCAGTGTGGTATTCAAAAAACTCTTTGATGGGCGAAAG

FIGURE 19-1

CAAGAGGATGTAGTCGAAATGAACGACAACTTTGAATTTGGACTCTGCCCATGT
GATGCCAAACCCATAGTAAGAGGGAAGTTCAATACAACGCTGCTGAACGGACC
GGCCTTCCAGATGGTATGCCCCATAGGATGGACAGGGACTGTAAGCTGTACGT
CATTCAATATGGACACCTTAGCCACAACTGTGGTACGGACATATAGAAGGTCTA
AACCATTCCCTCATAGGCAAGGCTGTATCACCCAAAAGAATCTGGGGGAGGAT
CTCCATAACTGCATCCTTGGAGGAAATTGGACTTGTGTGCCTGGAGACCAACTA
CTATACAAAGGGGGCTCTATTGAATCTTGCAAGTGGTGTGGCTATCAATTTAAA
GAGAGTGAGGGACTACCACACTACCCCATTGGCAAGTGTAAATTGGAGAACGA
GACTGGTTACAGGCTAGTAGACAGTACCTCTTGCAATAGAGAAGGTGTGGCCA
TAGTACCACAAGGGACATTAAAGTGCAAGATAGGAAAAACAACTGTACAGGTC
ATAGCTATGGATACCAAACTCGGACCTATGCCTTGCAGACCATATGAAATCATA
TCAAGTGAGGGGCCTGTAGAAAAGACAGCGTGTACTTTCAACTACACTAAGAC
ATTAAAAAATAAGTATTTTGAGCCCAGAGACAGCTACTTTCAGCAATACATGCT
AAAAGGAGAGTATCAATACTGGTTTGACCTGGAGGTGACTGACCATCACCGGG
ATTACTTCGCTGAGTCCATATTAGTGGTGGTAGTAGCCCTCTTGGGTGGCAGAT
ATGTACTTTGGTTACTGGTTACATACATGGTCTTATCAGAACAGAAGGCCTTAG
GGATTCAGTATGGATCAGGGGAAGTGGTGATGATGGGCAACTTGCTAACCCAT
AACAATATTGAAGTGGTGACATACTTCTTGCTGCTGTACCTACTGCTGAGGGAG
GAGAGCGTAAAGAAGTGGGTCTTACTCTTATACCACATCTTAGTGGTACACCCA
ATCAAATCTGTAATTGTGATCCTACTGATGATTGGGGATGTGGTAAAGGCCGAT
TCAGGGGGCCAAGAGTACTTGGGGAAAATAGACCTCTGTTTTACAACAGTAGT
ACTAATCGTCATAGGTTTAATCATAGCCAGGCGTGACCCAACTATAGTGCCACT
GGTAACAATAATGGCAGCACTGAGGGTCACTGAACTGACCCACCAGCCTGGAG
TTGACATCGCTGTGGCGGTCATGACTATAACCCTACTGATGGTTAGCTATGTGA
CAGATTATTTTAGATATAAAAAATGGTTACAGTGCATTCTCAGCCTGGTATCTGC
GGTGTTCTTGATAAGAAGCCTAATATACCTAGGTAGAATCGAGATGCCAGAGG
TAACTATCCCAAACTGGAGACCACTAACTTTAATACTATTATATTTGATCTCAAC
AACAATTGTAACGAGGTGGAAGGTTGACGTGGCTGGCCTATTGTTGCAATGTG
TGCCTATCTTATTGCTGGTCACAACCTTGTGGGCCGACTTCTTAACCCTAATACT
GATCCTGCCTACCTATGAATTGGTTAAATTATACTATCTGAAAACTGTTAGGACT
GATATAGAAAGAAGTTGGCTAGGGGGGATAGACTATACAAGAGTTGACTCCAT
CTACGACGTTGATGAGAGTGGAGAGGGCGTATATCTTTTTCCATCAAGGCAGA
AAGCACAGGGGAATTTTTCTATACTCTTGCCCCTTATCAAAGCAACACTGATAA
GTTGCGTCAGCAGTAAATGGCAGCTAATATACATGAGTTACTTAACTTTGGACT
TTATGTACTACATGCACAGGAAAGTTATAGAAGAGATCTCAGGAGGTACCAACA
TAATATCCAGGTTAGTGGCAGCACTCATAGAGCTGAACTGGTCCATGGAAGAA
GAGGAGAGCAAAGGCTTAAAGAAGTTTTATCTATTGTCTGGAAGGTTGAGAAA
CCTAATAATAAAACATAAGGTAAGGAATGAGACCGTGGCTTCTTGGTACGGGG
AGGAGGAAGTCTACGGTATGCCAAAGATCATGACTATAATCAAGGCCAGTACA
CTGAGTAAGAGCAGGCACTGCATAATATGCACTGTATGTGAGGGCCGAGAGTG
GAAAGGTGGCACCTGCCCAAAATGTGGACGCCATGGGAAGCCGATAACGTGT
GGGATGTCGCTAGCAGATTTTGAAGAAAGACACTATAAAAGAATCTTTATAAGG
GAAGGCAACTTTGAGGGTATGTGCAGCCGATGCCAGGGAAAGCATAGGAGGT
TTGAAATGGACCGGGAACCTAAGAGTGCCAGATACTGTGCTGAGTGTAATAGG
CTGCATCCTGCTGAGGAAGGTGACTTTTGGGCAGAGTCGAGCATGTTGGGCCT
CAAAATCACCTACTTTGCGCTGATGGATGGAAAGGTGTATGATATCACAGAGTG
GGCTGGATGCCAGCGTGTGGGAATCTCCCCAGATACCCACAGAGTCCCTTGTC
ACATCTCATTTGGTTCACGGATGCCTTTCAGGCAGGAATACAATGGCTTTGTAC
AATATACCGCTAGGGGGCAACTATTTCTGAGAAACTTGCCCGTACTGGCAACTA
AAGTAAAAAATGCTCATGGTAGGCAACCTTGGAGAAGAAATTGGTAATCTGGAA
CATCTTGGGTGGATCCTAAGGGGGCCTGCCGTGTGTAAGAAGATCACAGAGCA
CGAAAAATGCCACATTAATATACTGGATAAACTAACCGCATTTTTCGGGATCAT
GCCAAGGGGGACTACACCCAGAGCCCCGGTGAGGTTCCCTACGAGCTTACTAA
AAGTGAGGAGGGGTCTGGAGACTGCCTGGGCTTACACACACCAAGGCGGGAT

FIGURE 19-2

AAGTTCAGTCGACCATGTAACCGCCGGAAAAGATCTACTGGTCTGTGACAGCA
TGGGACGAACTAGAGTGGTTTGCCAAAGCAACAACAGGTTGACCGATGAGACA
GAGTATGGCGTCAAGACTGACTCAGGGTGCCCAGACGGTGCCAGATGTTATGT
GTTAAATCCAGAGGCCGTTAACATATCAGGATCCAAAGGGGCAGTCGTTCACC
TCCAAAAGACAGGTGGAGAATTCACGTGTGTCACCGCATCAGGCACACCGGCT
TTCTTCGACCTAAAAAACTTGAAAGGATGGTCAGGCTTGCCTATATTTGAAGCC
TCCAGCGGGAGGGTGGTTGGCAGAGTCAAAGTAGGGAAGAATGAAGAGTCTA
AACCTACAAAAATAATGAGTGGAATCCAGACCGTCTCAAAAAACAGAGCAGAC
CTGACCGAGATGGTCAAGAAGATAACCAGCATGAACAGGGGAGACTTCAAGCA
GATTACTTTGGCAACAGGGGCAGGCAAAACCACAGAACTCCCAAAAGCAGTTA
TAGAGGAGATAGGAAGACACAAGAGAGTATTAGTTCTTATACCATTAAGGGCA
GCGGCAGAGTCAGTCTACCAGTATATGAGATTGAAACACCCAAGCATCTCTTTT
AACCTAAGGATAGGGGACATGAAAGAGGGGGACATGGCAACCGGGATAACCT
ATGCATCATACGGGTACTTCTGCCAAATGCCTCAACCAAAGCTCAGAGCTGCTA
TGGTAGAATACTCATACATATTCTTAGATGAATACCATTGTGCCACTCCTGAACA
ACTGGCAATTATCGGGAAGATCCACAGATTTTCAGAGAGTATAAGGGTTGTCG
CCATGACTGCCACGCCAGCAGGGTCGGTGACCACAACAGGTCAAAAGCACCCA
ATAGAGGAATTCATAGCCCCCGAGGTAATGAAAGGGGAGGATCTTGGTAGTCA
GTTCCTTGATATAGCAGGGTTAAAAATACCAGTGGATGAGATGAAAGGCAATAT
GTTGGTTTTTGTACCAACGAGAAACATGGCAGTAGAGGTAGCAAAGAAGCTAA
AAGCTAAGGGCTATAACTCTGGATACTATTACAGTGGAGAGGATCCAGCCAAT
CTGAGAGTTGTGACATCACAATCCCCCTATGTAATCGTGGCTACAAATGCTATT
GAATCAGGAGTGACACTACCAGATTTGGACACGGTTATAGACACGGGGTTGAA
ATGTGAAAAGAGGGTGAGGGTATCATCAAAGATACCCTTCATCGTAACAGGCC
TTAAGAGGATGGCCGTGACTGTGGGTGAGCAGGCGCAGCGTAGGGGCAGAGT
AGGTAGAGTGAAACCCGGGAGGTATTATAGGAGCCAGGAAACAGCAACAGGG
TCAAAGGACTACCACTATGACCTCTTGCAGGCACAAAGATACGGGATTGAGGA
TGGAATCAACGTGACGAAATCCTTTAGGGAGATGAATTACGATTGGAGCCTATA
CGAGGAGGACAGCCTACTAATAACCCAGCTGGAAATACTAAATAATCTACTCAT
CTCAGAAGACTTGCCAGCCGCTGTTAAGAACATAATGGCCAGGACTGATCACC
CAGAGCCAATCCAACTTGCATACAACAGCTATGAAGTCCAGGTCCCGGTCCTGT
TCCCAAAAATAAGGAATGGAGAAGTCACAGACACCTACGAAAATTACTCGTTTC
TAAATGCCAGAAAGTTAGGGGAGGATGTGCCCGTGTATATCTACGCTACTGAA
GATGAGGATCTGGCAGTTGACCTCTTAGGGCTAGACTGGCCTGATCCTGGGAA
CCAGCAGGTAGTGGAGACTGGTAAAGCACTGAAGCAAGTGACCGGGTTGTCCT
CGGCTGAAAATGCCCTACTAGTGGCTTTATTTGGGTATGTGGGTTACCAGGCTC
TCTCAAAGAGGCATGTCCCAATGATAACAGACATATATACCATCGAGGACCAGA
GACTAGAAGACACCACCCACCTCCAGTATGCACCCAACGCCATAAAAACCGAT
GGGACAGAGACTGAACTGAAAGAACTGGCGTCGGGTGACGTGGAAAAAATCA
TGGGAGCCATTTCAGATTATGCAGCTGGGGGACTGGAGTTTGTTAAATCCCAA
GCAGAAAAGATAAAAACAGCTCCTTTGTTTAAAGAAAACGCAGAAGCCGCAAA
AGGGTATGTCCAAAAATTCATTGACTCATTAATTGAAAATAAAGAAGAAATAAT
CAGATATGGTTTGTGGGGAACACACACAGCACTATACAAAAGCATAGCTGCAA
GACTGGGGCATGAAACAGCGTTTGCCACACTAGTGTTAAAGTGGCTAGCTTTT
GGAGGGGAATCAGTGTCAGACCACGTCAAGCAGGCGGCAGTTGATTTAGTGG
TCTATTATGTGATGAATAAGCCTTCCTTCCCAGGTGACTCCGAGACACAGCAAG
AAGGGAGGCGATTCGTCGCAAGCCTGTTCATCTCCGCACTGGCAACCTACACA
TACAAAACTTGGAATTACCACAATCTCTCTAAAGTGGTGGAACCAGCCCTGGCT
TACCTCCCCTATGCTACCAGCGCATTAAAAATGTTCACCCCAACGCGGCTGGAG
AGCGTGGTGATACTGAGCACCACGATATATAAAACATACCTCTCTATAAGGAAG
GGGAAGAGTGATGGATTGCTGGGTACGGGGATAAGTGCAGCCATGGAAATCC
TGTCACAAAACCCAGTATCGGTAGGTATATCTGTGATGTTGGGGGTAGGGGCA
ATCGCTGCGCACAACGCTATTGAGTCCAGTGAACAGAAAAGGACCCTACTTAT
GAAGGTGTTTGTAAAGAACTTCTTGGATCAGGCTGCAACAGATGAGCTGGTAA

FIGURE 19-3

AAGAAAACCCAGAAAAAATTATAATGGCCTTATTTGAAGCAGTCCAGACAATTG
GTAACCCCCTGAGACTAATATACCACCTGTATGGGGTTTACTACAAAGGTTGGG
AGGCCAAGGAACTATCTGAGAGGACAGCAGGCAGAAACTTATTCACATTGATA
ATGTTTGAAGCCTTCGAGTTATTAGGGATGGACTCACAAGGGAAAATAAGGAA
CCTGTCCGGAAATTACATTTTGGATTTGATATACGGCCTACACAAGCAAATCAA
CAGAGGGCTGAAGAAAATGGTACTGGGGTGGGCCCCTGCACCCTTTAGTTGTG
ACTGGACCCCTAGTGACGAGAGGATCAGATTGCCAACAGACAACTATTTGAGG
GTAGAAACCAGGTGCCCATGTGGCTATGAGATGAAAGCTTTCAAAAATGTAGG
TGGCAAACTTACCAAAGTGGAGGAGAGCGGGCCTTTCCTATGTAGAAACAGAC
CTGGTAGGGGACCAGTCAACTACAGAGTCACCAAGTATTACGATGACAACCTC
AGAGAGATAAAACCAGTAGCAAAGTTGGAAGGACAGGTAGAGCACTACTACAA
AGGGGTCACAGCAAAAATTGACTACAGTAAAGGAAAAATGCTCTTGGCCACTG
ACAAGTGGGAGGTGGAACATGGTGTCATAACCAGGTTAGCTAAGAGATATACT
GGGGTCGGGTTCAATGGTGCATACTTAGGTGACGAGCCCAATCACCGTGCTCT
AGTGGAGAGGGACTGTGCAACTATAACCAAAAACACAGTACAGTTTCTAAAAAT
GAAGAAGGGGTGTGCGTTCACCTATGACCTGACCATCTCCAATCTGACCAGGC
TCATCGAACTAGTACACAGGAACAATCTTGAAGAGAAGGAAATACCCACCGCT
ACGGTCACCACATGGCTAGCTTACACCTTCGTGAATGAAGACGTAGGGACTAT
AAAACCAGTACTAGGAGAGAGAGTAATCCCCGACCCTGTAGTTGATATCAATTT
ACAACCAGAGGTGCAAGTGGACACGTCAGAGGTTGGGATCACAATAATTGGAA
GGGAAACCCTGATGACAACGGGAGTGACACCTGTCTTGGAAAAAGTAGAGCCT
GACGCCAGCGACAACCAAAACTCGGTGAAGATCGGGTTGGATGAGGGTAATTA
CCCAGGGCCTGGAATACAGACACATACACTAACAGAAGAAATACACAACAGGG
ATGCGAGGCCCTTCATCATGATCCTGGGCTCAAGGAATTCCATATCAAATAGGG
CAAAGACTGCTAGAAATATAAATCTGTACACAGGAAATGACCCCAGGGAAATA
CGAGACTTGATGGCTGCAGGGCGCATGTTAGTAGTAGCACTGAGGGATGTCGA
CCCTGAGCTGTCTGAAATGGTCGATTTCAAGGGGACTTTTTTAGATAGGGAGG
CCCTGGAGGCTCTAAGTCTCGGGCAACCTAAACCGAAGCAGGTTACCAAGGAA
GCTGTTAGGAATTTGATAGAACAGAAAAAAGATGTGGAGATCCCTAACTGGTTT
GCATCAGATGACCCAGTATTTCTGGAAGTGGCCTTAAAAAATGATAAGTACTAC
TTAGTAGGAGATGTTGGAGAGCTAAAAGATCAAGCTAAAGCACTTGGGGCCAC
GGATCAGACAAGAATTATAAAGGAGGTAGGCTCAAGGACGTATGCCATGAAGC
TATCTAGCTGGTTCCTCAAGGCATCAAACAAACAGATGAGTTTAACTCCACTGT
TTGAGGAATTGTTGCTACGGTGCCCACCTGCAACTAAGAGCAATAAGGGGCAC
ATGGCATCAGCTTACCAATTGGCACAGGGTAACTGGGAGCCCCTCGGTTGCGG
GGTGCACCTAGGTACAATACCAGCCAGAAGGGTGAAGATACACCCATATGAAG
CTTACCTGAAGTTGAAAGATTTCATAGAAGAAGAAGAGAAGAAACCTAGGGTT
AAGGATACAGTAATAAGAGAGCACAACAAATGGATACTTAAAAAAATAAGGTTT
CAAGGAAACCTCAACACCAAGAAAATGCTCAACCCGGGGAAACTATCTGAACA
GTTGGACAGGGAGGGGCGCAAGAGGAACATCTACAACCACCAGATTGGTACT
ATAATGTCAAGTGCAGGCATAAGGCTGGAGAAATTGCCAATAGTGAGGGCCCA
AACCGACACCAAAACCTTTCATGAGGCAATAAGAGATAAGATAGACAAGAGTG
AAAACCGGCAAAATCCAGAATTGCACAACAAATTGTTGGAGATTTTCCACACGA
TAGCCCAACCCACCCTGAAACACACCTACGGTGAGGTGACGTGGGAGCAACTT
GAGGCGGGGATAAATAGAAAGGGGGCAGCAGGCTTCCTGGAGAAGAAGAACA
TCGGAGAAGTATTGGATTCAGAAAAGCACCTGGTAGAACAATTGGTCAGGGAT
CTGAAGGCCGGGAGAAAGATAAAATATTATGAAACTGCAATACCAAAAAATGA
GAAGAGAGATGTCAGTGATGACTGGCAGGCAGGGGACCTGGTGGTTGAGAAG
AGGCCAAGAGTTATCCAATACCCTGAAGCCAAGACAAGGCTAGCCATCACTAA
GGTCATGTATAACTGGGTGAAACAGCAGCCCGTTGTGATTCCAGGATATGAAG
GAAAGACCCCCTTGTTCAACATCTTTGATAAAGTGAGAAAGGAATGGGACTCGT
TCAATGAGCCAGTGGCCGTAAGTTTTGACACCAAAGCCTGGGACACTCAAGTG
ACTAGTAAGGATCTGCAACTTATTGGAGAAATCCAGAAATATTACTATAAGAAG
GAGTGGCACAAGTTCATTGACACCATCACCGACCACATGACAGAAGTACCAGT

FIGURE 19-4

TATAACAGCAGATGGTGAAGTATATATAAGAAATGGGCAGAGAGGGAGCGGC
CAGCCAGACACAAGTGCTGGCAACAGCATGTTAAATGTCCTGACAATGATGTA
CGGCTTCTGCGAAAGCACAGGGGTACCGTACAAGAGTTTCAACAGGGTGGCAA
GGATCCACGTCTGTGGGGATGATGGCTTCTTAATAACTGAAAAAGGGTTAGGG
CTGAAATTTGCTAACAAAGGGATGCAGATTCTTCATGAAGCAGGCAAACCTCAG
AAGATAACGGAAGGGGAAAAGATGAAAGTTGCCTATAGATTTGAGGATATAGA
GTTCTGTTCTCATACCCCAGTCCCTGTTAGGTGGTCCGACAACACCAGTAGTCA
CATGGCCGGGAGAGACACCGCTGTGATACTATCAAAGATGGCAACAAGATTGG
ATTCAAGTGGAGAGAGGGGTACCACAGCATATGAAAAAGCGGTAGCCTTCAGT
TTCTTGCTGATGTATTCCTGGAACCCGCTTGTTAGGAGGATTTGCCTGTTGGTC
CTTTCGCAACAGCCAGAGACAGACCCATCAAAACATGCCACTTATTATTACAAA
GGTGATCCAATAGGGGCCTATAAAGATGTAATAGGTCGGAATCTAAGTGAACT
GAAGAGAACAGGCTTTGAGAAATTGGCAAATCTAAACCTAAGCCTGTCCACGTT
GGGGATCTGGACTAAGCACACAAGCAAAAGAATAATTCAGGACTGTGTTGCCA
TTGGGAAAGAAGAGGGCAACTGGCTAGTTAACGCCGACAGGCTGATATCCAGC
AAAACTGGCCACTTATACATACCTGATAAAGGCTTTACATTACAAGGAAAGCAT
TATGAGCAACTGCAGCTAAGAACAGAGACAAACCCGGTCATGGGGGTTGGGA
CTGAGAGATACAAGTTAGGTCCCATAGTCAATCTGCTGCTGAGAAGGTTGAAA
ATTCTGCTCATGACGGCCGTCGGCGTCAGCAGCTGAaggttggggtaaacactccggcctcttag
gccatttcctgtttttttttttttttttttttttttttttttttttttttttctttttttttttttttttccttttttttttttttttttttcttccttctttttcctttctttc
cttccttctttaatggtggctccatcttagccctagtcacggctagctgtgaaaggtccgtgagccgcatgactgcagagagtgctgatact
ggcctctctgcagatcatgtCCCCCGGCCGTCGGCGTCAGCTGAgacaaaatgtatatattgtaaataaattaatc
catgtacatagtgtatataaatatagttgggaccgtccacctcaagaagacgacacgcccaacacgcacagctaaacagtagtcaagatt
atctacctcaagataacactacatttaatgcacacagcactttagctgtatgaggatacgcccgacgtctatagttggactagggaagacct
ctaacagccccc

FIGURE 19-5

```
           AATTCTGCTCATGACGGCCGTCGGCGTCAGCAGCTGAAGGTTGGGGTAAACACTCCGGCCTCTTAGGCCA
                    10        20        30        40        50        60        70
3H3Bfrag   AATTCTGCTCATGACGGCCGTCGGCGTCAGCAGCTGAAGGTTGGGGTAAACACTCCGGCCTCTTAGGCCA 70
1.1.4 seq  AATTCTGCTCATGACGGCCGTCGGCGTCAGCAGCTGAAGGTTGGGGTAAACACTCCGGCCTCTTAGGCCA 70
1.2.3 seq  AATTCTGCTCATGACGGCCGTCGGCGTCAGCAGCTGAAGGTTGGGGTAAACACTCCGGCCTCTTAGGCCA 70
6.2.2 seq  AATTCTGCTCATGACGGCCGTCGGCGTCAGCAGCTGAAGGTTGGGGTAAACACTCCGGCCTCTTAGGCCA 70
6.1.4 seq  AATTCTGCTCATGACGGCCGTCGGCGTCAGCAGCTGAAGGTTGGGGTAAACACTCCGGCCTCTTAGGCCA 70

           TTTCCTGTTTTTTTTTTTTTTTTTTTTTTTTT--------------------------------------
                    80        90       100       110       120       130       140
3H3Bfrag   TTTCCTGTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTCTTTTTTTTTTT 140
1.1.4 seq  TTTCCTGTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTT------------------------------- 109
1.2.3 seq  TTTCCTGTTTTTTTTTTTTTTTTTTTTTTTTT-------------------------------------- 102
6.2.2 seq  TTTCCTGTTTTTTTTTTTTTTTTTTTTTT----------------------------------------- 99
6.1.4 seq  TTTCCTGTTTTTTT-------------------------------------------------------- 84

           ----------------------------CTTTCCTTCTTTTTTTCCTTTCTTTTCCTTCCTTCTTTAATG
                   150       160       170       180       190       200       210
3H3Bfrag   TTTTTTCCTTTTTTTTTTTTTTTTTTTTCTTTCCTTCTTTNTTTCCTTTCTTTTCCTTCCTTCTTTAATG 210
1.1.4 seq  ----------------------------CTTTCCTTCTTTTTT-CCTTTCTTTTCCTTCCTTCTTTAATG 149
1.2.3 seq  ----------------------------CTTTCCTTCTTTTTT-CCTTTCTTTTCCTTCCTTCTTTAATG 142
6.2.2 seq  ----------------------------CTTTCCTTCTTTTTTTCCTTTCTTTTCCTTCCTTCTTTAATG 140
6.1.4 seq  ----------------------------CTTTCCTTCTTTTTTTCCTTTCTTTTCCTTCCTTCTTTAATG 125

           GTGGCTCCATCTTAGCCCTAGTCACGGCTAGCTGTGAAAGGTCCGTGAGCCGCATGACTGCAGAGAGTGC
                   220       230       240       250       260       270       280
3H3Bfrag   GTGGCTCCATCTTAGCCCTAGTCACGGCTAGCTGTGAAAGGTCCGTGAGCCGCATGACTGCAGAGAGTGC 280
1.1.4 seq  GTGGCTCCATCTTAGCCCTAGTCACGGCTAGCTGTGAAAGGTCCGTGAGCCGCATGACTGCAGAGAGTGC 219
1.2.3 seq  GTGGCTCCATCTTAGCCCTAGTCACGGCTAGCTGTGAAAGGTCCGTGAGCCGCATGACTGCAGAGAGTGC 212
6.2.2 seq  GTGGCTCCATCTTAGCCCTAGTCACGGCTAGCTGTGAAAGGTCCGTGAGCCGCATGACTGCAGAGAGTGC 210
6.1.4 seq  GTGGCTCCATCTTAGCCCTAGTCACGGCTAGCTGTGAAAGGTCCGTGAGCCGCATGACTGCAGAGAGTGC 195

           TGATACTGGCCTCTCTGCAGATCATGTCCCCCGGCCGTCGGCGTCAGCAGCTGAGACAAAATGTATATAT
                   290       300       310       320       330       340       350
3H3Bfrag   TGATACTGGCCTCTCTGCAGATCATGTCCCCCGGCCGTCGGCGTCAGCAGCTGAGACAAAATGTATATAT 350
1.1.4 seq  TGATACTGGCCTCTCTGCAGATCATGTCCCCCGGCCGTCGGCGTCAGCAGCTGAGACAAAATGTATATAT 289
1.2.3 seq  TGATACTGGCCTCTCTGCAGATCATGTCCCCCGGCCGTCGGCGTCAGCAGCTGAGACAAAATGTATATAT 282
6.2.2 seq  TGATACTGGCCTCTCTGCAGATCATGTCCCCCGGCCGTCGGCGTCAGCAGCTGAGACAAAATGTATATAT 280
6.1.4 seq  TGATACTGGCCTCTCTGCAGATCATGTCCCCCGGCCGTCGGCGTCAGCAGCTGAGACAAAATGTATATAT 265

           TGTAAATAAATTAATCCATGTACATAGTGTATATAAATATAGTTGGGACCGT
                   360       370       380       390       400
3H3Bfrag   TGTAAATAAATTAATCCATGTACATAGTGTATATAAATATAGTTGGGACCGT 402
1.1.4 seq  TGTAAATAAATTAATCCATGTACATAGTGTATATAAATATAGTTGGGACCGT 341
1.2.3 seq  TGTAAATAAATTAATCCATGTACATAGTGTATATAAATATAGTTGGGACCGT 334
6.2.2 seq  TGTAAATAAATTAATCCATGTACATAGTGTATATAAATATAGTTGGGACCGT 332
6.1.4 seq  TGTAAATAAATTAATCCATGTACATAGTGTATATAAATATAGTTGGGACCGT 317
```

FIGURE 20

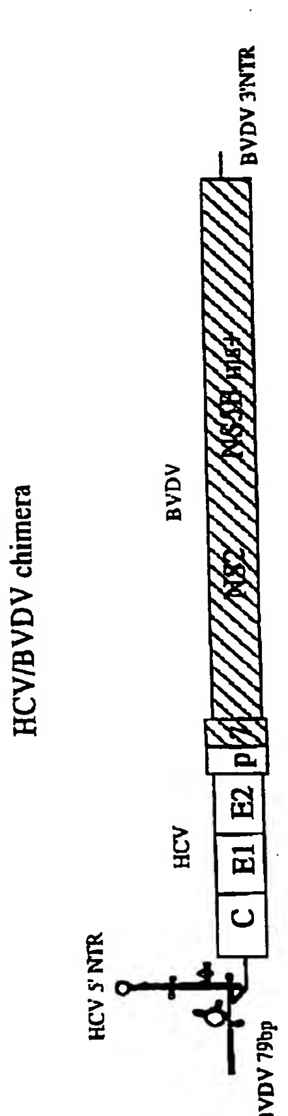
HCV/BVDV chimera
HCV
BVDV
HCV 5' NTR
BVDV 79bp
C
E1
E2
P
NS2
NS5B
BVDV 3'NTR

FIGURE 21

Gtatacgagaattagaaaaggcactcgtatacgtattgggcaattaaaaataataattaggcctaggtacatggcacgtgccagccccct
gatgggggcgacactccaccatgaatcactcccctgtgaggaactactgtcttcacgcagaaagcgtctagccatggcgttagtatgag
tgtcgtgcagcctccaggaccccccctcccgggagagccatagtggtctgcggaaccggtgagtacaccggaattgccaggacgac
cgggtcctttcttggataaacccgctcaatgcctggagatttgggcgtgcccccgcaagactgctagccgagtagtgttgggtcgcgaa
aggccttgtggtactgcctgatagggtgcttgcgagtgccccgggaggtctcgtagaccgtgcaccATGAGCACGAATC
CTAAACCTCAAAGAAAAACCAAACGTAACACCAACCGTCGCCCACAGGACGTC
AAGTTCCCGGGTGGCGGTCAGATCGTTGGTGGAGTTTACTTGTTGCCGCGCAG
GGGCCCTAGATTGGGTGTGCGCGCGACGAGGAAGACTTCCGAGCGGTCGCAA
CCTCGAGGTAGACGTCAGCCTATCCCCAAGGCACGTCGGCCCGAGGGCAGGA
CCTGGGCTCAGCCCGGGTACCCTTGGCCCCTCTATGGCAATGAGGGTTGCGGG
TGGGCGGGATGGCTCCTGTCTCCCCGTGGCTCTCGGCCTAGCTGGGGCCCCAC
AGACCCCCGGCGTAGGTCGCGCAATTTGGGTAAGGTCATCGATACCCTTACGT
GCGGCTTCGCCGACCTCATGGGGTACATACCGCTCGTCGGCGCCCCTCTTGGA
GGCGCTGCCAGGGCCCTGGCGCATGGCGTCCGGGTTCTGGAAGACGGCGTGA
ACTATGCAACAGGGAACCTTCCTGGTTGCTCTTTCTCTATCTTCCTTCTGGCCCT
GCTCTCTTGCCTGACCGTGCCCGCTTCAGCCTACCAAGTGCGCAATTCCTCGGG
GCTTTACCATGTCACCAATGATTGCCCTAACTCGAGTATTGTGTACGAGGCGGC
CGATGCCATCCTGCACACTCCGGGGTGTGTCCCTTGCGTTCGCGAGGGTAACG
CCTCGAGGTGTTGGGTGGCGGTGACCCCCACGGTGGCCACCAGGGACGGCAA
ACTCCCCACAACGCAGCTTCGACGTCATATCGATCTGCTTGTCGGGAGCGCCA
CCCTCTGCTCGGCCCTCTACGTGGGGGACCTGTGCGGGTCTGTCTTTCTTGTTG
GTCAACTGTTTACCTTCTCTCCCAGGCGCCACTGGACGACGCAAGACTGCAATT
GTTCTATCTATCCCGGCCATATAACGGGTCATCGCATGGCATGGGATATGATGA
TGAACTGGTCCCCTACGGCAGCGTTGGTGGTAGCTCAGCTGCTCCGGATCCCA
CAAGCCATCATGGACATGATCGCTGGTGCTCACTGGGGAGTCCTGGCGGGCAT
AGCGTATTTCTCCATGGTGGGGAACTGGGCGAAGGTCCTGGTAGTGCTGCTGC
TATTTGCCGGCGTCGACGCGGAAACCCACGTCACCGGGGGAAGTGCCGGCCG
CACCACGGCTGGGCTTGTTGGTCTCCTTACACCAGGCGCCAAGCAGAACATCC
AACTGATCAACACCAACGGCAGTTGGCACATCAATAGCACGGCCTTGAACTGC
AATGAAAGCCTTAACACCGGCTGGTTAGCAGGGCTCTTCTATCAGCACAAATTC
AACTCTTCAGGCTGTCCTGAGAGGTTGGCCAGCTGCCGACGCCTTACCGATTTT
GCCCAGGGCTGGGGTCCTATCAGTTATGCCAACGGAAGCGGCCTCGACGAAC
GCCCCTACTGCTGGCACTACCCTCCAAGACCTTGTGGCATTGTGCCCGCAAAG
AGCGTGTGTGGCCCGGTATATTGCTTCACTCCCAGCCCCGTGGTGGTGGGAAC
GACCGACAGGTCGGGCGCGCCTACCTACAGCTGGGGTGCAAATGATACGGAT
GTCTTCGTCCTTAACAACACCAGGCCACCGCTGGGCAATTGGTTCGGTTGTACC
TGGATGAACTCAACTGGATTCACCAAAGTGTGCGGAGCGCCCCCTTGTGTCAT
CGGAGGGGTGGGCAACAACACCTTGCTCTGCCCCACTGATTGTTTCCGCAAGC
ATCCGGAAGCCACATACTCTCGGTGCGGCTCCGGTCCCTGGATTACACCCAGG
TGCATGGTCGACTACCCGTATAGGCTTTGGCACTATCCTTGTACCATCAATTAC
ACCATATTCAAAGTCAGGATGTACGTGGGAGGGGTCGAGCACAGGCTGGAAG
CGGCCTGCAACTGGACGCGGGGCGAACGCTGTGATCTGGAAGACAGGGACAG
GTCCGAGCTCAGCCCATTGCTGCTGTCCACCACACAGTGGCAGGTCCTTCCGT
GTTCTTTCACGACCCTGCCAGCCTTGTCCACCGGCCTCATCCACCTCCACCAGA
ACATTGTGGACGTGCAGTACTTGTACGGGGTAGGGTCAAGCATCGCGTCCTGG
GCCATTAAGTGGGAGTACGTCGTTCTCCTGTTCCTCCTGCTTGCAGACGCGCGC
GTCTGCTCCTGCTTGTGGATGATGTTACTCATATCCCAAGCGGAGGCGGCTTTG
GAGAACCTCGTAATACTCAATGCAGCATCCCTGGCCGGGACGCACGGTCTTGT
GTCCTTCCTCGTGTTCTTCTGCTTTGCGTGGTATCTGAAGGGTAGGTGGGTGCC

FIGURE 22-1

CGGAGCGGTCTACGCCTTCTACGGGAAGTGGGTCTTACTCTTATACCACATCTT
AGTGGTACACCCAATCAAATCTGTAATTGTGATCCTACTGATGATTGGGGATGT
GGTAAAGGCCGATTCAGGGGGCCAAGAGTACTTGGGGAAAATAGACCTCTGTT
TTACAACAGTAGTACTAATCGTCATAGGTTTAATCATAGCTAGGCGTGACCCAA
CTATAGTGCCACTGGTAACAATAATGGCAGCACTGAGGGTCACTGAACTGACC
CACCAGCCTGGAGTTGACATCGCTGTGGCGGTCATGACTATAACCCTACTGAT
GGTTAGCTATGTGACAGATTATTTTAGATATAAAAAATGGTTACAGTGCATTCTC
AGCCTGGTATCTGCGGTGTTCTTGATAAGAAGCCTAATATACCTAGGTAGAATC
GAGATGCCAGAGGTAACTATCCCAAACTGGAGACCACTAACTTTAATACTATTA
TATTTGATCTCAACAACAATTGTAACGAGGTGGAAGGTTGACGTGGCTGGCCTA
TTGTTGCAATGTGTGCCTATCTTATTGCTGGTCACAACCTTGTGGGCCGACTTCT
TAACCCTAATACTGATCCTGCCTACCTATGAATTGGTTAAATTATACTATCTGAA
AACTGTTAGGACTGATACAGAAAGAAGTTGGCTAGGGGGGATAGACTATACAA
GAGTTGACTCCATCTACGACGTTGATGAGAGTGGAGAGGGCGTATATCTTTTTC
CATCAAGGCAGAAAGCACAGGGGAATTTTTCTATACTCTTGCCCCTTATCAAAG
CAACACTGATAAGTTGCGTCAGCAGTAAATGGCAGCTAATATACATGAGTTACT
TAACTTTGGACTTTATGTACTACATGCACAGGAAAGTTATAGAAGAGATCTCAG
GAGGTACCAACATAATATCCAGGTTAGTGGCAGCACTCATAGAGCTGAACTGG
TCCATGGAAGAAGAGGAGAGCAAAGGCTTAAAGAAGTTTTATCTATTGTCTGG
AAGGTTGAGAAACCTAATAATAAAACATAAGGTAAGGAATGAGACCGTGGCTT
CTTGGTACGGGGAGGAGGAAGTCTACGGTATGCCAAAGATCATGACTATAATC
AAGGCCAGTACACTGAGTAAGAGCAGGCACTGCATAATATGCACTGTATGTGA
GGGCCGAGAGTGGAAAGGTGGCACCTGCCCAAAATGTGGACGCCATGGGAAG
CCGATAACGTGTGGGATGTCGCTAGCAGATTTTGAAGAAAGACACTATAAAAG
AATCTTTATAAGGGAAGGCAACTTTGAGGGTATGTGCAGCCGATGCCAGGGAA
AGCATAGGAGGTTTGAAATGGACCGGGAACCTAAGAGTGCCAGATACTGTGCT
GAGTGTAATAGGCTGCATCCTGCTGAGGAAGGTGACTTTTGGGCAGAGTCGAG
CATGTTGGGCCTCAAAATCACCTACTTTGCGCTGATGGATGGAAAGGTGTATGA
TATCACAGAGTGGGCTGGATGCCAGCGTGTGGGAATCTCCCCAGATACCCACA
GAGTCCCTTGTCACATCTCATTTGGTTCACGGATGCCTTTCAGGCAGGAATACA
ATGGCTTTGTACAATATACCGCTAGGGGGCAACTATTTCTGAGAAACTTGCCCG
TACTGGCAACTAAAGTAAAAATGCTCATGGTAGGCAACCTTGGAGAAGAAATT
GGTAATCTGGAACATCTTGGGTGGATCCTAAGGGGGCCTGCCGTGTGTAAGAA
GATCACAGAGCACGAAAAATGCCACATTAATATACTGGATAAACTAACCGCATT
TTTCGGGATCATGCCAAGGGGGACTACACCCAGAGCCCCGGTGAGGTTCCCTA
CGAGCTTACTAAAAGTGAGGAGGGGTCTGGAGACTGCCTGGGCTTACACACAC
CAAGGCGGGATAAGTTCAGTCGACCATGTAACCGCCGGAAAAGATCTACTGGT
CTGTGACAGCATGGGACGAACTAGAGTGGTTTGCCAAAGCAACAACAGGTTGA
CCGATGAGACAGAGTATGGCGTCAAGACTGACTCAGGGTGCCCAGACGGTGC
CAGATGTTATGTGTTAAATCCAGAGGCCGTTAACATATCAGGATCCAAAGGGG
CAGTCGTTCACCTCCAAAAGACAGGTGGAGAATTCACGTGTGTCACCGCATCA
GGCACACCGGCTTTCTTCGACCTAAAAAACTTGAAAGGATGGTCAGGCTTGCCT
ATATTTGAAGCCTCCAGCGGGAGGGTGGTTGGCAGAGTCAAAGTAGGGAAGA
ATGAAGAGTCTAAACCTACAAAAATAATGAGTGGAATCCAGACCGTCTCAAAAA
ACAGAGCAGACCTGACCGAGATGGTCAAGAAGATAACCAGCATGAACAGGGG
AGACTTCAAGCAGATTACTTTGGCAACAGGGGCAGGCAAAACCACAGAACTCC
CAAAAGCAGTTATAGAGGAGATAGGAAGACACAAGAGAGTATTAGTTCTTATA
CCATTAAGGGCAGCGGCAGAGTCAGTCTACCAGTATATGAGATTGAAACACCC
AAGCATCTCTTTTAACCTAAGGATAGGGGACATGAAAGAGGGGGACATGGCAA
CCGGGATAACCTATGCATCATACGGGTACTTCTGCCAAATGCCTCAACCAAAGC
TCAGAGCTGCTATGGTAGAATACTCATACATATTCTTAGATGAATACCATTGTGC
CACTCCTGAACAACTGGCAATTATCGGGAAGATCCACAGATTTTCAGAGAGTAT
AAGGGTTGTCGCCATGACTGCCACGCCAGCAGGGTCGGTGACCACAACAGGT
CAAAAGCACCCAATAGAGGAATTCATAGCCCCCGAGGTAATGAAAGGGGAGG

FIGURE 22-2

ATCTTGGTAGTCAGTTCCTTGATATAGCAGGGTTAAAAATACCAGTGGATGAGA
TGAAAGGCAATATGTTGGTTTTTGTACCAACGAGAAACATGGCAGTAGAGGTA
GCAAAGAAGCTAAAAGCTAAGGGCTATAACTCTGGATACTATTACAGTGGAGA
GGATCCAGCCAATCTGAGAGTTGTGACATCACAATCCCCCTATGTAATCGTGGC
TACAAATGCTATTGAATCAGGAGTGACACTACCAGATTTGGACACGGTTATAGA
CACGGGGTTGAAATGTGAAAAGAGGGTGAGGGTATCATCAAAGATACCCTTCA
TCGTAACAGGCCTTAAGAGGATGGCCGTGACTGTGGGTGAGCAGGCGCAGCG
TAGGGGCAGAGTAGGTAGAGTGAAACCCGGGAGGTATTATAGGAGCCAGGAA
ACAGCAACAGGGTCAAAGGACTACCACTATGACCTCTTGCAGGCACAAAGATA
CGGGATTGAGGATGGAATCAACGTGACGAAATCCTTTAGGGAGATGAATTACG
ATTGGAGCCTATACGAGGAGGACAGCCTACTAATAACCCAGCTGGAAATACTA
AATAATCTACTCATCTCAGAAGACTTGCCAGCCGCTGTTAAGAACATAATGGCC
AGGACTGATCACCCAGAGCCAATCCAACTTGCATACAACAGCTATGAAGTCCA
GGTCCCGGTCCTATTCCCAAAAATAAGGAATGGAGAAGTCACAGACACCTACG
AAAATTACTCGTTTCTAAATGCCAGAAAGTTAGGGGAGGATGTGCCCGTGTATA
TCTACGCTACTGAAGATGAGGATCTGGCAGTTGACCTCTTAGGGCTAGACTGG
CCTGATCCTGGGAACCAGCAGGTAGTGGAGACTGGTAAAGCACTGAAGCAAGT
GACCGGGTTGTCCTCGGCTGAAAATGCCCTACTAGTGGCTTTATTTGGGTATGT
GGGTTACCAGGCTCTCTCAAAGAGGCATGTCCCAATGATAACAGACATATATAC
CATCGAGGACCAGAGACTAGAAGACACCACCCACCTCCAGTATGCACCCAACG
CCATAAAAACCGATGGGACAGAGACTGAACTGAAAGAACTGGCGTCGGGTGA
CGTGGAAAAAATCATGGGAGCCATTTCAGATTATGCAGCTGGGGGACTGGAGT
TTGTTAAATCCCAAGCAGAAAAGATAAAAACAGCTCCTTTGTTTAAAGAAAACG
CAGAAGCCGCAAAAGGGTATGTCCAAAAATTCATTGACTCATTAATTGAAAATA
AAGAAGAAATAATCAGATATGGTTTGTGGGGAACACACACAGCACTATACAAA
AGCATAGCTGCAAGACTGGGGCATGAAACAGCGTTTGCCACACTAGTGTTAAA
GTGGCTAGCTTTTGGAGGGGAATCAGTGTCAGACCACGTCAAGCAGGCGGCA
GTTGATTTAGTGGTCTATTATGTGATGAATAAGCCTTCCTTCCCAGGTGACTCC
GAGACACAGCAAGAAGGGAGGCGATTCGTCGCAAGCCTGTTCATCTCCGCACT
GGCAACCTACACATACAAAACTTGGAATTACCACAATCTCTCTAAAGTGGTGGA
ACCAGCCCTGGCTTACCTCCCCTATGCTACCAGCGCATTAAAAATGTTCACCCC
AACGCGGCTGGAGAGCGTGGTGATACTGAGCACCACGATATATAAAACATACC
TCTCTATAAGGAAGGGGAAGAGTGATGGATTGCTGGGTACGGGGATAAGTGC
AGCCATGGAAATCCTGTCACAAAACCCAGTATCGGTAGGTATATCTGTGATGTT
GGGGGTAGGGGCAATCGCTGCGCACAACGCTATTGAGTCCAGTGAACAGAAA
AGGACCCTACTTATGAAGGTGTTTGTAAAGAACTTCTTGGATCAGGCTGCAACA
GATGAGCTGGTAAAAGAAAACCCAGAAAAAATTATAATGGCCTTATTTGAAGCA
GTCCAGACAATTGGTAACCCCCTGAGACTAATATACCACCTGTATGGGGTTTAC
TACAAAGGTTGGGAGGCCAAGGAACTATCTGAGAGGACAGCAGGCAGAAACT
TATTCACATTGATAATGTTTGAAGCCTTCGAGTTATTAGGGATGGACTCACAAG
GGAAAATAAGGAACCTGTCCGGAAATTACATTTTGGATTTGATATACGGCCTAC
ACAAGCAAATCAACAGAGGGCTGAAGAAAATGGTACTGGGGTGGGCCCCTGC
ACCCTTTAGTTGTGACTGGACCCCTAGTGACGAGAGGATCAGATTGCCAACAG
ACAACTATTTGAGGGTAGAAACCAGGTGCCCATGTGGCTATGAGATGAAAGCT
TTCAAAAATGTAGGTGGCAAACTTACCAAAGTGGAGGAGAGCGGGCCTTTCCT
ATGTAGAAACAGACCTGGTAGGGGACCAGTCAACTACAGAGTCACCAAGTATT
ACGATGACAACCTCAGAGAGATAAAACCAGTAGCAAAGTTGGAAGGACAGGTA
GAGCACTACTACAAAGGGGTCACAGCAAAAATTGACTACAGTAAAGGAAAAAT
GCTCTTGGCCACTGACAAGTGGGAGGTGGAACATGGTGTCATAACCAGGTTAG
CTAAGAGATATACTGGGGTCGGGTTCAATGGTGCATACTTAGGTGACGAGCCC
AATCACCGTGCTCTAGTGGAGAGGGACTGTGCAACTATAACCAAAAACACAGT
ACAGTTTCTAAAAATGAAGAAGGGGTGTGCGTTCACCTATGACCTGACCATCTC
CAATCTGACCAGGCTCATCGAACTAGTACACAGGAACAATCTTGAAGAGAAGG
AAATACCCACCGCTACGGTCACCACATGGCTAGCTTACACCTTCGTGAATGAAG

FIGURE 22-3

ACGTAGGGACTATAAAACCAGTACTAGGAGAGAGAGTAATCCCCGACCCTGTA
GTTGATATCAATTTACAACCAGAGGTGCAAGTGGACACGTCAGAGGTTGGGAT
CACAATAATTGGAAGGGAAACCCTGATGACAACGGGAGTGACACCTGTCTTGG
AAAAAGTAGAGCCTGACGCCAGCGACAACCAAAACTCGGTGAAGATCGGGTTG
GATGAGGGTAATTACCCAGGGCCTGGAATACAGACACATACACTAACAGAAGA
AATACACAACAGGGATGCGAGGCCCTTCATCATGATCCTGGGCTCAAGGAATT
CCATATCAAATAGGGCAAAGACTGCTAGAAATATAAATCTGTACACAGGAAATG
ACCCCAGGGAAATACGAGACTTGATGGCTGCAGGGCGCATGTTAGTAGTAGCA
CTGAGGGATGTCGACCCTGAGCTGTCTGAAATGGTCGATTTCAAGGGGACTTT
TTTAGATAGGGAGGCCCTGGAGGCTCTAAGTCTCGGGCAACCTAAACCGAAGC
AGGTTACCAAGGAAGCTGTTAGGAATTTGATAGAACAGAAAAAAGATGTGGAG
ATCCCTAACTGGTTTGCATCAGATGACCCAGTATTTCTGGAAGTGGCCTTAAAA
AATGATAAGTACTACTTAGTAGGAGATGTTGGAGAGCTAAAAGATCAAGCTAAA
GCACTTGGGGCCACGGATCAGACAAGAATTATAAAGGAGGTAGGCTCAAGGA
CGTATGCCATGAAGCTATCTAGCTGGTTCCTCAAGGCATCAAACAAACAGATGA
GTTTAACTCCACTGTTTGAGGAATTGTTGCTACGGTGCCCACCTGCAACTAAGA
GCAATAAGGGGCACATGGCATCAGCTTACCAATTGGCACAGGGTAACTGGGAG
CCCCTCGGTTGCGGGGTGCACCTAGGTACAATACCAGCCAGAAGGGTGAAGAT
ACACCCATATGAAGCTTACCTGAAGTTGAAAGATTTCATAGAAGAAGAAGAGAA
GAAACCTAGGGTTAAGGATACAGTAATAAGAGAGCACAACAAATGGATACTTA
AAAAAATAAGGTTTCAAGGAAACCTCAACACCAAGAAAATGCTCAACCCAGGG
AAACTATCTGAACAGTTGGACAGGGAGGGGCGCAAGAGGAACATCTACAACCA
CCAGATTGGTACTATAATGTCAAGTGCAGGCATAAGGCTGGAGAAATTGCCAA
TAGTGAGGGCCCAAACCGACACCAAAACCTTTCATGAGGCAATAAGAGATAAG
ATAGACAAGAGTGAAAACCGGCAAAATCCAGAATTGCACAACAAATTGTTGGA
GATTTTCCACACGATAGCCCAACCCACCCTGAAACACACCTACGGTGAGGTGA
CGTGGGAGCAACTTGAGGCGGGGGTAAATAGAAAGGGGGCAGCAGGCTTCCT
GGAGAAGAAGAACATCGGAGAAGTATTGGATTCAGAAAAGCACCTGGTAGAAC
AATTGGTCAGGGATCTGAAGGCCGGGAGAAAGATAAAATATTATGAAACTGCA
ATACCAAAAAATGAGAAGAGAGATGTCAGTGATGACTGGCAGGCAGGGGACC
TGGTGGTTGAGAAGAGGCCAAGAGTTATCCAATACCCTGAAGCCAAGACAAGG
CTAGCCATCACTAAGGTCATGTATAACTGGGTGAAACAGCAGCCCGTTGTGATT
CCAGGATATGAAGGAAAGACCCCCTTGTTCAACATCTTTGATAAAGTGAGAAAG
GAATGGGACTCGTTCAATGAGCCAGTGGCCGTAAGTTTTGACACCAAAGCCTG
GGACACTCAAGTGACTAGTAAGGATCTGCAACTTATTGGAGAAATCCAGAAATA
TTACTATAAGAAGGAGTGGCACAAGTTCATTGACACCATCACCGACCACATGAC
AGAAGTACCAGTTATAACAGCAGATGGTGAAGTATATATAAGAAATGGGCAGA
GAGGGAGCGGCCAGCCAGACACAAGTGCTGGCAACAGCATGTTAAATGTCCT
GACAATGATGTACGGCTTCTGCGAAAGCACAGGGGTACCGTACAAGAGTTTCA
ACAGGGTGGCAAGGATCCACGTCTGTGGGGATGATGGCTTCTTAATAACTGAA
AAAGGGTTAGGGCTGAAATTTGCTAACAAAGGGATGCAGATTCTTCATGAAGC
AGGCAAACCTCAGAAGATAACGGAAGGGGAAAAGATGAAAGTTGCCTATAGAT
TTGAGGATATAGAGTTCTGTTCTCATACCCCAGTCCCTGTTAGGTGGTCCGACA
ACACCAGTAGTCACATGGCCGGGAGAGACACCGCTGTGATACTATCAAAGATG
GCAACAAGATTGGATTCAAGTGGAGAGAGGGGTACCACAGCATATGAAAAAGC
GGTAGCCTTCAGTTTCTTGCTGATGTATTCCTGGAACCCGCTTGTTAGGAGGAT
TTGCCTGTTGGTCCTTTCGCAACAGCCAGAGACAGACCCATCAAAACATGCCAC
TTATTATTACAAAGGTGATCCAATAGGGGCCTATAAAGATGTAATAGGTCGGAA
TCTAAGTGAACTGAAGAGAACAGGCTTTGAGAAATTGGCAAATCTAAACCTAAG
CCTGTCCACGTTGGGGGTCTGGACTAAGCACACAAGCAAAAGAATAATTCAGG
ACTGTGTTGCCATTGGGAAAGAAGAGGGCAACTGGCTAGTTAAGCCCGACAGG
CTGATATCCAGCAAAACTGGCCACTTATACATACCTGATAAAGGCTTTACATTAC
AAGGAAAGCATTATGAGCAACTGCAGCTAAGAACAGAGACAAACCCGGTCATG
GGGGTTGGGACTGAGAGATACAAGTTAGGTCCCATAGTCAATCTGCTGCTGAG

FIGURE 22-4

AAGGTTGAAAATTCTGCTCATGACGGCCGTCGGCGTCAGCAGCTGAgacaaaatgtat
atattgtaaataaattaatccatgtacatagtgtatataaatatagttgggaccgtccacctcaagaagacgacacgcccaacacgcacag
ctaaacagtagtcaagattatctacctcaagataacactacatttaatgcacacagcactttagctgtatgaggatacgcccgacgtctatag
ttggactagggaagacctctaacagccccc

FIGURE 22-5

HCV/BVDV chimera with selectable marker

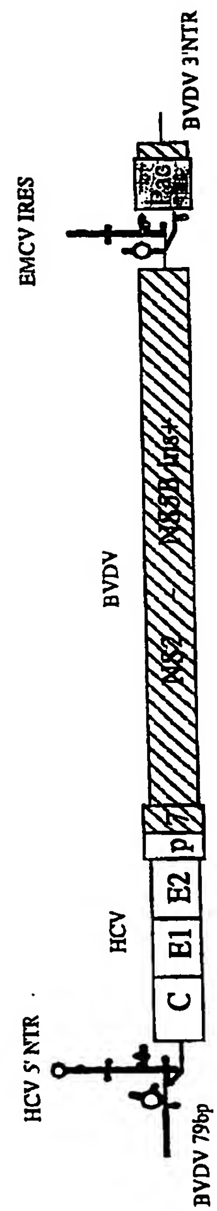

FIGURE 23

Gtatacgagaattagaaaaggcactcgtatacgtattgggcaattaaaaataataattaggcctaggtacatggcacgtgccagcccccct
gatgggggcgacactccaccatgaatcactcccctgtgaggaactactgtcttcacgcagaaagcgtctagccatggcgttagtatgag
tgtcgtgcagcctccaggacccccccctcccgggagagccatagtggtctgcggaaccggtgagtacaccggaattgccaggacgac
cgggtcctttcttggataaacccgctcaatgcctggagatttgggcgtgcccccgcaagactgctagccgagtagtgttgggtcgcgaa
aggccttgtggtactgcctgatagggtgcttgcgagtgccccgggaggtctcgtagaccgtgcaccATGAGCACGAATC
CTAAACCTCAAAGAAAAACCAAACGTAACACCAACCGTCGCCCACAGGACGTC
AAGTTCCCGGGTGGCGGTCAGATCGTTGGTGGAGTTTACTTGTTGCCGCGCAG
GGGCCCTAGATTGGGTGTGCGCGCGACGAGGAAGACTTCCGAGCGGTCGCAA
CCTCGAGGTAGACGTCAGCCTATCCCCAAGGCACGTCGGCCCGAGGGCAGGA
CCTGGGCTCAGCCCGGGTACCCTTGGCCCCTCTATGGCAATGAGGGTTGCGGG
TGGGCGGGATGGCTCCTGTCTCCCCGTGGCTCTCGGCCTAGCTGGGGCCCCAC
AGACCCCCGGCGTAGGTCGCGCAATTTGGGTAAGGTCATCGATACCCTTACGT
GCGGCTTCGCCGACCTCATGGGGTACATACCGCTCGTCGGCGCCCCTCTTGGA
GGCGCTGCCAGGGCCCTGGCGCATGGCGTCCGGGTTCTGGAAGACGGCGTGA
ACTATGCAACAGGGAACCTTCCTGGTTGCTCTTTCTCTATCTTCCTTCTGGCCCT
GCTCTCTTGCCTGACCGTGCCCGCTTCAGCCTACCAAGTGCGCAATTCCTCGGG
GCTTTACCATGTCACCAATGATTGCCCTAACTCGAGTATTGTGTACGAGGCGGC
CGATGCCATCCTGCACACTCCGGGGTGTGTCCCTTGCGTTCGCGAGGGTAACG
CCTCGAGGTGTTGGGTGGCGGTGACCCCCACGGTGGCCACCAGGGACGGCAA
ACTCCCCACAACGCAGCTTCGACGTCATATCGATCTGCTTGTCGGGAGCGCCA
CCCTCTGCTCGGCCCTCTACGTGGGGGACCTGTGCGGGTCTGTCTTTCTTGTTG
GTCAACTGTTTACCTTCTCTCCCAGGCGCCACTGGACGACGCAAGACTGCAATT
GTTCTATCTATCCCGGCCATATAACGGGTCATCGCATGGCATGGGATATGATGA
TGAACTGGTCCCCTACGGCAGCGTTGGTGGTAGCTCAGCTGCTCCGGATCCCA
CAAGCCATCATGGACATGATCGCTGGTGCTCACTGGGGAGTCCTGGCGGGCAT
AGCGTATTTCTCCATGGTGGGGAACTGGGCGAAGGTCCTGGTAGTGCTGCTGC
TATTTGCCGGCGTCGACGCGGAAACCCACGTCACCGGGGGAAGTGCCGGCCG
CACCACGGCTGGGCTTGTTGGTCTCCTTACACCAGGCGCCAAGCAGAACATCC
AACTGATCAACACCAACGGCAGTTGGCACATCAATAGCACGGCCTTGAACTGC
AATGAAAGCCTTAACACCGGCTGGTTAGCAGGGCTCTTCTATCAGCACAAATTC
AACTCTTCAGGCTGTCCTGAGAGGTTGGCCAGCTGCCGACGCCTTACCGATTTT
GCCCAGGGCTGGGGTCCTATCAGTTATGCCAACGGAAGCGGCCTCGACGAAC
GCCCCTACTGCTGGCACTACCCTCCAAGACCTTGTGGCATTGTGCCCGCAAAG
AGCGTGTGTGGCCCGGTATATTGCTTCACTCCCAGCCCCGTGGTGGTGGGAAC
GACCGACAGGTCGGGCGCGCCTACCTACAGCTGGGGTGCAAATGATACGGAT
GTCTTCGTCCTTAACAACACCAGGCCACCGCTGGGCAATTGGTTCGGTTGTACC
TGGATGAACTCAACTGGATTCACCAAAGTGTGCGGAGCGCCCCCTTGTGTCAT
CGGAGGGGTGGGCAACAACACCTTGCTCTGCCCCACTGATTGTTTCCGCAAGC
ATCCGGAAGCCACATACTCTCGGTGCGGCTCCGGTCCCTGGATTACACCCAGG
TGCATGGTCGACTACCCGTATAGGCTTTGGCACTATCCTTGTACCATCAATTAC
ACCATATTCAAAGTCAGGATGTACGTGGGAGGGGTCGAGCACAGGCTGGAAG
CGGCCTGCAACTGGACGCGGGGCGAACGCTGTGATCTGGAAGACAGGGACAG
GTCCGAGCTCAGCCCATTGCTGCTGTCCACCACACAGTGGCAGGTCCTTCCGT
GTTCTTTCACGACCCTGCCAGCCTTGTCCACCGGCCTCATCCACCTCCACCAGA
ACATTGTGGACGTGCAGTACTTGTACGGGGTAGGGTCAAGCATCGCGTCCTGG
GCCATTAAGTGGGAGTACGTCGTTCTCCTGTTCCTCCTGCTTGCAGACGCGCGC
GTCTGCTCCTGCTTGTGGATGATGTTACTCATATCCCAAGCGGAGGCGGCTTTG
GAGAACCTCGTAATACTCAATGCAGCATCCCTGGCCGGGACGCACGGTCTTGT

FIGURE 24-1

GTCCTTCCTCGTGTTCTTCTGCTTTGCGTGGTATCTGAAGGGTAGGTGGGTGCC
CGGAGCGGTCTACGCCTTCTACGGGAAGTGGGTCTTACTCTTATACCACATCTT
AGTGGTACACCCAATCAAATCTGTAATTGTGATCCTACTGATGATTGGGGATGT
GGTAAAGGCCGATTCAGGGGGCCAAGAGTACTTGGGGAAAATAGACCTCTGTT
TTACAACAGTAGTACTAATCGTCATAGGTTTAATCATAGCTAGGCGTGACCCAA
CTATAGTGCCACTGGTAACAATAATGGCAGCACTGAGGGTCACTGAACTGACC
CACCAGCCTGGAGTTGACATCGCTGTGGCGGTCATGACTATAACCCTACTGAT
GGTTAGCTATGTGACAGATTATTTTAGATATAAAAAATGGTTACAGTGCATTCTC
AGCCTGGTATCTGCGGTGTTCTTGATAAGAAGCCTAATATACCTAGGTAGAATC
GAGATGCCAGAGGTAACTATCCCAAACTGGAGACCACTAACTTTAATACTATTA
TATTTGATCTCAACAACAATTGTAACGAGGTGGAAGGTTGACGTGGCTGGCCTA
TTGTTGCAATGTGTGCCTATCTTATTGCTGGTCACAACCTTGTGGGCCGACTTCT
TAACCCTAATACTGATCCTGCCTACCTATGAATTGGTTAAATTATACTATCTGAA
AACTGTTAGGACTGATACAGAAAGAAGTTGGCTAGGGGGGATAGACTATACAA
GAGTTGACTCCATCTACGACGTTGATGAGAGTGGAGAGGGCGTATATCTTTTTC
CATCAAGGCAGAAAGCACAGGGGAATTTTTCTATACTCTTGCCCCTTATCAAAG
CAACACTGATAAGTTGCGTCAGCAGTAAATGGCAGCTAATATACATGAGTTACT
TAACTTTGGACTTTATGTACTACATGCACAGGAAAGTTATAGAAGAGATCTCAG
GAGGTACCAACATAATATCCAGGTTAGTGGCAGCACTCATAGAGCTGAACTGG
TCCATGGAAGAAGAGGAGAGCAAAGGCTTAAAGAAGTTTTATCTATTGTCTGG
AAGGTTGAGAAACCTAATAATAAAACATAAGGTAAGGAATGAGACCGTGGCTT
CTTGGTACGGGGAGGAGGAAGTCTACGGTATGCCAAAGATCATGACTATAATC
AAGGCCAGTACACTGAGTAAGAGCAGGCACTGCATAATATGCACTGTATGTGA
GGGCCGAGAGTGGAAAGGTGGCACCTGCCCAAAATGTGGACGCCATGGGAAG
CCGATAACGTGTGGGATGTCGCTAGCAGATTTTGAAGAAAGACACTATAAAAG
AATCTTTATAAGGGAAGGCAACTTTGAGGGTATGTGCAGCCGATGCCAGGGAA
AGCATAGGAGGTTTGAAATGGACCGGGAACCTAAGAGTGCCAGATACTGTGCT
GAGTGTAATAGGCTGCATCCTGCTGAGGAAGGTGACTTTTGGGCAGAGTCGAG
CATGTTGGGCCTCAAAATCACCTACTTTGCGCTGATGGATGGAAAGGTGTATGA
TATCACAGAGTGGGCTGGATGCCAGCGTGTGGGAATCTCCCCAGATACCCACA
GAGTCCCTTGTCACATCTCATTTGGTTCACGGATGCCTTTCAGGCAGGAATACA
ATGGCTTTGTACAATATACCGCTAGGGGGCAACTATTTCTGAGAAACTTGCCCG
TACTGGCAACTAAAGTAAAAATGCTCATGGTAGGCAACCTTGGAGAAGAAATT
GGTAATCTGGAACATCTTGGGTGGATCCTAAGGGGGCCTGCCGTGTGTAAGAA
GATCACAGAGCACGAAAAATGCCACATTAATATACTGGATAAACTAACCGCATT
TTTCGGGATCATGCCAAGGGGGACTACACCCAGAGCCCCGGTGAGGTTCCCTA
CGAGCTTACTAAAAGTGAGGAGGGGTCTGGAGACTGCCTGGGCTTACACACAC
CAAGGCGGGATAAGTTCAGTCGACCATGTAACCGCCGGAAAAGATCTACTGGT
CTGTGACAGCATGGGACGAACTAGAGTGGTTTGCCAAAGCAACAACAGGTTGA
CCGATGAGACAGAGTATGGCGTCAAGACTGACTCAGGGTGCCCAGACGGTGC
CAGATGTTATGTGTTAAATCCAGAGGCCGTTAACATATCAGGATCCAAAGGGG
CAGTCGTTCACCTCCAAAAGACAGGTGGAGAATTCACGTGTGTCACCGCATCA
GGCACACCGGCTTTCTTCGACCTAAAAAACTTGAAAGGATGGTCAGGCTTGCCT
ATATTTGAAGCCTCCAGCGGGAGGGTGGTTGGCAGAGTCAAAGTAGGGAAGA
ATGAAGAGTCTAAACCTACAAAAATAATGAGTGGAATCCAGACCGTCTCAAAAA
ACAGAGCAGACCTGACCGAGATGGTCAAGAAGATAACCAGCATGAACAGGGG
AGACTTCAAGCAGATTACTTTGGCAACAGGGGCAGGCAAAACCACAGAACTCC
CAAAAGCAGTTATAGAGGAGATAGGAAGACACAAGAGAGTATTAGTTCTTATA
CCATTAAGGGCAGCGGCAGAGTCAGTCTACCAGTATATGAGATTGAAACACCC
AAGCATCTCTTTTAACCTAAGGATAGGGGACATGAAAGAGGGGGACATGGCAA
CCGGGATAACCTATGCATCATACGGGTACTTCTGCCAAATGCCTCAACCAAAGC
TCAGAGCTGCTATGGTAGAATACTCATACATATTCTTAGATGAATACCATTGTGC
CACTCCTGAACAACTGGCAATTATCGGGAAGATCCACAGATTTTCAGAGAGTAT
AAGGGTTGTCGCCATGACTGCCACGCCAGCAGGGTCGGTGACCACAACAGGT

FIGURE 24-2

CAAAAGCACCCAATAGAGGAATTCATAGCCCCCGAGGTAATGAAAGGGGAGG
ATCTTGGTAGTCAGTTCCTTGATATAGCAGGGTTAAAAATACCAGTGGATGAGA
TGAAAGGCAATATGTTGGTTTTTGTACCAACGAGAAACATGGCAGTAGAGGTA
GCAAAGAAGCTAAAAGCTAAGGGCTATAACTCTGGATACTATTACAGTGGAGA
GGATCCAGCCAATCTGAGAGTTGTGACATCACAATCCCCCTATGTAATCGTGGC
TACAAATGCTATTGAATCAGGAGTGACACTACCAGATTTGGACACGGTTATAGA
CACGGGGTTGAAATGTGAAAAGAGGGTGAGGGTATCATCAAAGATACCCTTCA
TCGTAACAGGCCTTAAGAGGATGGCCGTGACTGTGGGTGAGCAGGCGCAGCG
TAGGGGCAGAGTAGGTAGAGTGAAACCCGGGAGGTATTATAGGAGCCAGGAA
ACAGCAACAGGGTCAAAGGACTACCACTATGACCTCTTGCAGGCACAAAGATA
CGGGATTGAGGATGGAATCAACGTGACGAAATCCTTTAGGGAGATGAATTACG
ATTGGAGCCTATACGAGGAGGACAGCCTACTAATAACCCAGCTGGAAATACTA
AATAATCTACTCATCTCAGAAGACTTGCCAGCCGCTGTTAAGAACATAATGGCC
AGGACTGATCACCCAGAGCCAATCCAACTTGCATACAACAGCTATGAAGTCCA
GGTCCCGGTCCTGTTCCCAAAAATAAGGAATGGAGAAGTCACAGACACCTACG
AAAATTACTCGTTTCTAAATGCCAGAAAGTTAGGGGAGGATGTGCCCGTGTATA
TCTACGCTACTGAAGATGAGGATCTGGCAGTTGACCTCTTAGGGCTAGACTGG
CCTGATCCTGGGAACCAGCAGGTAGTGGAGACTGGTAAAGCACTGAAGCAAGT
GACCGGGTTGTCCTCGGCTGAAAATGCCCTACTAGTGGCTTTATTTGGGTATGT
GGGTTACCAGGCTCTCTCAAAGAGGCATGTCCCAATGATAACAGACATATATAC
CATCGAGGACCAGAGACTAGAAGACACCACCCACCTCCAGTATGCACCCAACG
CCATAAAAACCGATGGGACAGAGACTGAACTGAAAGAACTGGCGTCGGGTGA
CGTGGAAAAAATCATGGGAGCCATTTCAGATTATGCAGCTGGGGGACTGGAGT
TTGTTAAATCCCAAGCAGAAAAGATAAAAACAGCTCCTTTGTTTAAAGAAAACG
CAGAAGCCGCAAAAGGGTATGTCCAAAAATTCATTGACTCATTAATTGAAAATA
AAGAAGAAATAATCAGATATGGTTTGTGGGGAACACACACAGCACTATACAAA
AGCATAGCTGCAAGACTGGGGCATGAAACAGCGTTTGCCACACTAGTGTTAAA
GTGGCTAGCTTTTGGAGGGGAATCAGTGTCAGACCACGTCAAGCAGGCGGCA
GTTGATTTAGTGGTCTATTATGTGATGAATAAGCCTTCCTTCCCAGGTGACTCC
GAGACACAGCAAGAAGGGAGGCGATTCGTCGCAAGCCTGTTCATCTCCGCACT
GGCAACCTACACATACAAAACTTGGAATTACCACAATCTCTCTAAAGTGGTGGA
ACCAGCCCTGGCTTACCTCCCCTATGCTACCAGCGCATTAAAAATGTTCACCCC
AACGCGGCTGGAGAGCGTGGTGATACTGAGCACCACGATATATAAAACATACC
TCTCTATAAGGAAGGGGAAGAGTGATGGATTGCTGGGTACGGGGATAAGTGC
AGCCATGGAAATCCTGTCACAAAACCCAGTATCGGTAGGTATATCTGTGATGTT
GGGGGTAGGGGCAATCGCTGCGCACAACGCTATTGAGTCCAGTGAACAGAAA
AGGACCCTACTTATGAAGGTGTTTGTAAAGAACTTCTTGGATCAGGCTGCAACA
GATGAGCTGGTAAAAGAAAACCCAGAAAAAATTATAATGGCCTTATTTGAAGCA
GTCCAGACAATTGGTAACCCCCTGAGACTAATATACCACCTGTATGGGGTTTAC
TACAAAGGTTGGGAGGCCAAGGAACTATCTGAGAGGACAGCAGGCAGAAACT
TATTCACATTGATAATGTTTGAAGCCTTCGAGTTATTAGGGATGGACTCACAAG
GGAAAATAAGGAACCTGTCCGGAAATTACATTTTGGATTTGATATACGGCCTAC
ACAAGCAAATCAACAGAGGGCTGAAGAAAATGGTACTGGGGTGGCCCCCTGC
ACCCTTTAGTTGTGACTGGACCCCTAGTGACGAGAGGATCAGATTGCCAACAG
ACAACTATTTGAGGGTAGAAACCAGGTGCCCATGTGGCTATGAGATGAAAGCT
TTCAAAAATGTAGGTGGCAAACTTACCAAAGTGGAGGAGAGCGGGCCTTTCCT
ATGTAGAAACAGACCTGGTAGGGGACCAGTCAACTACAGAGTCACCAAGTATT
ACGATGACAACCTCAGAGAGATAAAACCAGTAGCAAAGTTGGAAGGACAGGTA
GAGCACTACTACAAAGGGGTCACAGCAAAAATTGACTACAGTAAAGGAAAAAT
GCTCTTGGCCACTGACAAGTGGGAGGTGGAACATGGTGTCATAACCAGGTTAG
CTAAGAGATATACTGGGGTCGGGTTCAATGGTGCATACTTAGGTGACGAGCCC
AATCACCGTGCTCTAGTGGAGAGGGACTGTGCAACTATAACCAAAAACACAGT
ACAGTTTCTAAAAATGAAGAAGGGGTGTGCGTTCACCTATGACCTGACCATCTC
CAATCTGACCAGGCTCATCGAACTAGTACACAGGAACAATCTTGAAGAGAAGG

FIGURE 24-3

AAATACCCACCGCTACGGTCACCACATGGCTAGCTTACACCTTCGTGAATGAAG
ACGTAGGGACTATAAAACCAGTACTAGGAGAGAGAGTAATCCCCGACCCTGTA
GTTGATATCAATTTACAACCAGAGGTGCAAGTGGACACGTCAGAGGTTGGGAT
CACAATAATTGGAAGGGAAACCCTGATGACAACGGGAGTGACACCTGTCTTGG
AAAAAGTAGAGCCTGACGCCAGCGACAACCAAAACTCGGTGAAGATCGGGTTG
GATGAGGGTAATTACCCAGGGCCTGGAATACAGACACATACACTAACAGAAGA
AATACACAACAGGGATGCGAGGCCCTTCATCATGATCCTGGGCTCAAGGAATT
CCATATCAAATAGGGCAAAGACTGCTAGAAATATAAATCTGTACACAGGAAATG
ACCCCAGGGAAATACGAGACTTGATGGCTGCAGGGCGCATGTTAGTAGTAGCA
CTGAGGGATGTCGACCCTGAGCTGTCTGAAATGGTCGATTTCAAGGGGACTTT
TTTAGATAGGGAGGCCCTGGAGGCTCTAAGTCTCGGGCAACCTAAACCGAAGC
AGGTTACCAAGGAAGCTGTTAGGAATTTGATAGAACAGAAAAAAGATGTGGAG
ATCCCTAACTGGTTTGCATCAGATGACCCAGTATTTCTGGAAGTGGCCTTAAAA
AATGATAAGTACTACTTAGTAGGAGATGTTGGAGAGGTAAAAGATCAAGCTAA
AGCACTTGGGGCCACGGATCAGACAAGAATTATAAAGGAGGTAGGCTCAAGG
ACGTATGCCATGAAGCTATCTAGCTGGTTCCTCAAGGCATCAAACAAACAGATG
AGTTTAACTCCACTGTTTGAGGAATTGTTGCTACGGTGCCCACCTGCAACTAAG
AGCAATAAGGGGCACATGGCATCAGCTTACCAATTGGCACAGGGTAACTGGGA
GCCCCTCGGTTGCGGGGTGCACCTAGGTACAATACCAGCCAGAAGGGTGAAG
ATACACCCATATGAAGCTTACCTGAAGTTGAAAGATTTCATAGAAGAAGAAGAG
AAGAAACCTAGGGTTAAGGATACAGTAATAAGAGAGCACAACAAATGGATACT
TAAAAAAATAAGGTTTCAAGGAAACCTCAACACCAAGAAAATGCTCAACCCTGG
GAAACTATCTGAACAGTTGGACAGGGAGGGGCGCAAGAGGAACATCTACAAC
CACCAGATTGGTACTATAATGTCAAGTGCAGGCATAAGGCTGGAGAAATTGCC
AATAGTGAGGGCCCAAACCGACACCAAAACCTTTCATGAGGCAATAAGAGATA
AGATAGACAAGAGTGAAAACCGGCAAAATCCAGAATTGCACAACAAATTGTTG
GAGATTTTCCACACGATAGCCCAACCCACCCTGAAACACACCTACGGTGAGGT
GACGTGGGAGCAACTTGAGGCGGGGATAAATAGAAAGGGGGCAGCAGGCTTC
CTGGAGAAGAAGAACATCGGAGAAGTATTGGATTCAGAAAAGCACCTGGTAGA
ACAATTGGTCAGGGATCTGAAGGCCGGGAGAAAGATAAAATATTATGAAACTG
CAATACCAAAAAATGAGAAGAGAGATGTCAGTGATGACTGGCAGGCAGGGGA
CCTGGTGGTTGAGAAGAGGCCAAGAGTTATCCAATACCCTGAAGCCAAGACAA
GGCTAGCCATCACTAAGGTCATGTATAACTGGGTGAAACAGCAGCCCGTTGTG
ATTCCAGGATATGAAGGAAAGACCCCCTTGTTCAACATCTTTGATAAAGTGAGA
AAGGAATGGGACTCGTTCAATGAGCCAGTGGCCGTAAGTTTTGACACCAAAGC
CTGGGACACTCAAGTGACTAGTAAGGATCTGCAACTTATTGGAGAAATCCAGA
AATATTACTATAAGAAGGAGTGGCACAAGTTCATTGACACCATCACCGACCACA
TGACAGAAGTACCAGTTATAACAGCAGATGGTGAAGTATATATAAGAAATGGG
CAGAGAGGGAGCGGCCAGCCAGACACAAGTGCTGGCAACAGCATGTTAAATG
TCCTGACAATGATGTACGCCTTCTGCGAAAGCACAGGGGTACCGTACAAGAGT
TTCAACAGGGTGGCAAGGATCCACGTCTGTGGGGATGATGGCTTCTTAATAAC
TGAAAAAGGGTTAGGGCTGAAATTTGCTAACAAAGGGATGCAGATTCTTCATG
AAGCAGGCAAACCTCAGAAGATAACGGAAGGGGAAAAGATGAAAGTTGCCTAT
AGATTTGAGGATATAGAGTTCTGTTCTCATACCCCAGTCCCTGTTAGGTGGTCC
GACAACACCAGTAGTCACATGGCCGGGAGAGACACCGCTGTGATACTATCAAA
GATGGCAACAAGATTGGATTCAAGTGGAGAGAGGGGTACCACAGCATATGAAA
AAGCGGTAGCCTTCAGTTTCTTGCTGATGTATTCCTGGAACCCGCTTGTTAGGA
GGATTTGCCTGTTGGTCCTTTCGCAACAGCCAGAGACAGACCCATCAAAACATG
CCACTTATTATTACAAAGGTGATCCAATAGGGGCCTATAAAGATGTAATAGGTC
GGAATCTAAGTGAACTGAAGAGAACAGGCTTTGAGAAATTGGCAAATCTAAAC
CTAAGCCTGTCCACGTTGGGGATCTGGACTAAGCACACAAGCAAAAGAATAAT
TCAGGACTGTGTTGCCATTGGGAAAGAAGAGGGCAACTGGCTAGTTAACGCCG
ACAGGCTGATATCCAGCAAAACTGGCCACTTATACATACCTGATAAAGGCTTTA
CATTACAAGGAAAGCATTATGAGCAACTGCAGCTAAGAACAGAGACAAACCCG

FIGURE 24-4

GTCATGGGGGTTGGGACTGAGAGATACAAGTTAGGTCCCATAGTCAATCTGCT
GCTGAGAAGGTTGAAAATTCTGCTCATGACGGCCGTCGGCGTCAGCAGCTGAg
acaaaatgtatatattgtaaataaattaatccatgtacAATTCCGCCCCTCTCCCTCCCCCCCCCCTAACG
TTACTGGCCGAAGCCGCTTGGAATAAGGCCGGTGTGCGTTTGTCTATATGTTAT
TTTCCACCATATTGCCGTCTTTTGGCAATGTGAGGGCCCGGAAACCTGGCCCTG
TCTTCTTGACGAGCATTCCTAGGGGTCTTTCCCCTCTCGCCAAAGGAATGCAAG
GTCTGTTGAATGTCGTGAAGGAAGCAGTTCCTCTGGAAGCTTCTTGAAGACAAA
CAACGTCTGTAGCGACCCTTTGCAGGCAGCGGAACCCCCCACCTGGCGACAGG
TGCCTCTGCGGCCAAAAGCCACGTGTATAAGATACACCTGCAAAGGCGGCACA
ACCCCAGTGCCACGTTGTGAGTTGGATAGTTGTGGAAAGAGTCAAATGGCTCT
CCTCAAGCGTATTCAACAAGGGGCTGAAGGATGCCCAGAAGGTACCCCATTGT
ATGGGATCTGATCTGGGGCCTCGGTGCACATGCTTTACATGTGTTTAGTCGAG
GTTAAAAAACGTCTAGGCCCCCCGAACCACGGGGACGTGGTTTTCCTTTGAAA
AACACGATGATAAGCTTGCCACAACcatgaccgagtacaagcccacggtgcgcctcgccacccgcgacga
cgtccccgggccgtacgcaccctcgccgccgcgttcgccgactaccccgccacgcgccacaccgtcgacccggaccgccacatc
gagcgggtcaccgagctgcaagaactcttcctcacgcgcgtcgggctcgacatcggcaaggtgtgggtcgcggacgacggcgcc
gcggtggcggtctggaccacgccggagagcgtcgaagcgggggcggtgttcgccgagatcggcccgcgcatggccgagttgag
cggttcccggctggccgcgcagcaacagatggaaggcctcctggcgccgcaccggcccaaggagcccgcgtggttcctggccac
cgtcggcgtctcgcccgaccaccagggcaagggtctgggcagcgccgtcgtgctccccggagtggaggcggccgagcgcgccg
gggtgcccgccttcctggagacctccgcgccccgcaacctccccttctacgagcggctcggcttcaccgtcaccgccgacgtcgagt
gcccgaaggaccgcgcgacctggtgcatgacccgcaagcccggtgccTGAcgcccgccccacgacccgcagcgcccgaccg
aaaggagcgcacgaccccatgaaATGCATCGATCGTACGAATTAACGCCGACAGGCTGATAT
CCAGCAAAACTGGCCACTTATACATACCTGATAAAGGCTTTACATTACAAGGAA
AGCATTATGAGCAACTGCAGCTAAGAACAGAGACAAACCCGGTCATGGGGGTT
GGGACTGAGAGATACAAGTTAGGTCCCATAGTCAATCTGCTGCTGAGAAGGTT
GAAAATTCTGCTCATGACGGCCGTCGGCGTCAGCAGCTGAgacaaaatgtatatattgtaaata
aattaatccatgtacatagtgtatataaatatagttgggaccgtccacctcaagaagacgacacgcccaacacgcacagctaaacagtag
tcaagattatctacctcaagataacactacatttaatgcacacagcactttagctgtatgaggatacgcccgacgtctatagttggactagg
gaagacctctaacagccccc

FIGURE 24-5

Bicistronic HCV/BVDV chimera

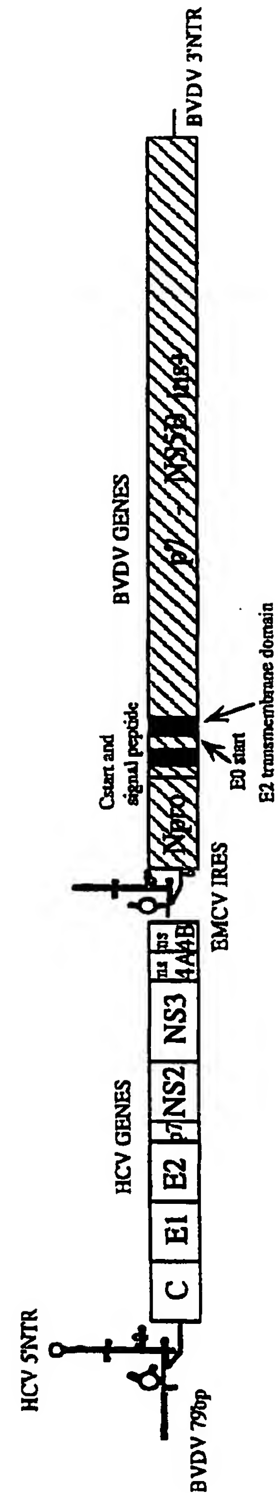

FIGURE 25

Gtatacgagaattagaaaaggcactcgtatacgtattgggcaattaaaaataataattaggcctaggtacatggcacgtgccagcccccct
gatgggggcgacactccaccatgaatcactcccctgtgaggaactactgtcttcacgcagaaagcgtctagccatggcgttagtatgag
tgtcgtgcagcctccaggaccccccctcccgggagagccatagtggtctgcggaaccggtgagtacaccggaattgccaggacgac
cgggtcctttcttggataaacccgctcaatgcctggagatttgggcgtgcccccgcaagactgctagccgagtagtgttgggtcgcgaa
aggccttgtggtactgcctgatagggtgcttgcgagtgccccgggaggtctcgtagaccgtgcaccATGAGCACGAATC
CTAAACCTCAAAGAAAAACCAAACGTAACACCAACCGTCGCCCACAGGACGTC
AAGTTCCCGGGTGGCGGTCAGATCGTTGGTGGAGTTTACTTGTTGCCGCGCAG
GGGCCCTAGATTGGGTGTGCGCGCGACGAGGAAGACTTCCGAGCGGTCGCAA
CCTCGAGGTAGACGTCAGCCTATCCCCAAGGCACGTCGGCCCGAGGGCAGGA
CCTGGGCTCAGCCCGGGTACCCTTGGCCCCTCTATGGCAATGAGGGTTGCGGG
TGGGCGGGATGGCTCCTGTCTCCCCGTGGCTCTCGGCCTAGCTGGGGCCCCAC
AGACCCCCGGCGTAGGTCGCGCAATTTGGGTAAGGTCATCGATACCCTTACGT
GCGGCTTCGCCGACCTCATGGGGTACATACCGCTCGTCGGCGCCCCTCTTGGA
GGCGCTGCCAGGGCCCTGGCGCATGGCGTCCGGGTTCTGGAAGACGGCGTGA
ACTATGCAACAGGGAACCTTCCTGGTTGCTCTTTCTCTATCTTCCTTCTGGCCCT
GCTCTCTTGCCTGACCGTGCCCGCTTCAGCCTACCAAGTGCGCAATTCCTCGGG
GCTTTACCATGTCACCAATGATTGCCCTAACTCGAGTATTGTGTACGAGGCGGC
CGATGCCATCCTGCACACTCCGGGGTGTGTCCCTTGCGTTCGCGAGGGTAACG
CCTCGAGGTGTTGGGTGGCGGTGACCCCCACGGTGGCCACCAGGGACGGCAA
ACTCCCCACAACGCAGCTTCGACGTCATATCGATCTGCTTGTCGGGAGCGCCA
CCCTCTGCTCGGCCCTCTACGTGGGGGACCTGTGCGGGTCTGTCTTTCTTGTTG
GTCAACTGTTTACCTTCTCTCCCAGGCGCCACTGGACGACGCAAGACTGCAATT
GTTCTATCTATCCCGGCCATATAACGGGTCATCGCATGGCATGGGATATGATGA
TGAACTGGTCCCCTACGGCAGCGTTGGTGGTAGCTCAGCTGCTCCGGATCCCA
CAAGCCATCATGGACATGATCGCTGGTGCTCACTGGGGAGTCCTGGCGGGCAT
AGCGTATTTCTCCATGGTGGGGAACTGGGCGAAGGTCCTGGTAGTGCTGCTGC
TATTTGCCGGCGTCGACGCGGAAACCCACGTCACCGGGGGAAGTGCCGGCCG
CACCACGGCTGGGCTTGTTGGTCTCCTTACACCAGGCGCCAAGCAGAACATCC
AACTGATCAACACCAACGGCAGTTGGCACATCAATAGCACGGCCTTGAACTGC
AATGAAAGCCTTAACACCGGCTGGTTAGCAGGGCTCTTCTATCAGCACAAATTC
AACTCTTCAGGCTGTCCTGAGAGGTTGGCCAGCTGCCGACGCCTTACCGATTTT
GCCCAGGGCTGGGGTCCTATCAGTTATGCCAACGGAAGCGGCCTCGACGAAC
GCCCCTACTGCTGGCACTACCCTCCAAGACCTTGTGGCATTGTGCCCGCAAAG
AGCGTGTGTGGCCCGGTATATTGCTTCACTCCCAGCCCCGTGGTGGTGGGAAC
GACCGACAGGTCGGGCGCGCCTACCTACAGCTGGGGTGCAAATGATACGGAT
GTCTTCGTCCTTAACAACACCAGGCCACCGCTGGGCAATTGGTTCGGTTGTACC
TGGATGAACTCAACTGGATTCACCAAAGTGTGCGGAGCGCCCCCTTGTGTCAT
CGGAGGGGTGGGCAACAACACCTTGCTCTGCCCCACTGATTGTTTCCGCAAGC
ATCCGGAAGCCACATACTCTCGGTGCGGCTCCGGTCCCTGGATTACACCCAGG
TGCATGGTCGACTACCCGTATAGGCTTTGGCACTATCCTTGTACCATCAATTAC
ACCATATTCAAAGTCAGGATGTACGTGGGAGGGGTCGAGCACAGGCTGGAAG
CGGCCTGCAACTGGACGCGGGGCGAACGCTGTGATCTGGAAGACAGGGACAG
GTCCGAGCTCAGCCCATTGCTGCTGTCCACCACACAGTGGCAGGTCCTTCCGT
GTTCTTTCACGACCCTGCCAGCCTTGTCCACCGGCCTCATCCACCTCCACCAGA
ACATTGTGGACGTGCAGTACTTGTACGGGGTAGGGTCAAGCATCGCGTCCTGG
GCCATTAAGTGGGAGTACGTCGTTCTCCTGTTCCTCCTGCTTGCAGACGCGCGC
GTCTGCTCCTGCTTGTGGATGATGTTACTCATATCCCAAGCGGAGGCGGCTTTG
GAGAACCTCGTAATACTCAATGCAGCATCCCTGGCCGGGACGCACGGTCTTGT
GTCCTTCCTCGTGTTCTTCTGCTTTGCGTGGTATCTGAAGGGTAGGTGGGTGCC

FIGURE 26-1

CGGAGCGGTCTACGCCTTCTACGGGATGTGGCCTCTCCTCCTGCTCCTGCTGG
CGTTGCCTCAGCGGGCATACGCACTGGACACGGAGGTGGCCGCGTCGTGTGG
CGGCGTTGTTCTTGTCGGGTTAATGGCGCTGACTCTGTCGCCATATTACAAGCG
CTACATCAGCTGGTGCATGTGGTGGCTTCAGTATTTTCTGACCAGAGTAGAAGC
GCAACTGCACGTGTGGGTTCCCCCCCTCAACGTCCGGGGGGGGCGCGATGCC
GTCATCTTACTCATGTGTGTTGTACACCCGACTCTGGTATTTGACATCACCAAAC
TACTCCTGGCCATCTTCGGACCCCTTTGGATTCTTCAAGCCAGTTTGCTTAAAGT
CCCCTACTTCGTGCGCGTTCAAGGCCTTCTCCGGATCTGCGCGCTAGCGCGGA
AGATAGCCGGAGGTCATTACGTGCAAATGGCCATCATCAAGTTAGGGGCGCTT
ACTGGCACCTATGTGTATAACCATCTCACCCCTCTTCGAGACTGGGCGCACAAC
GGCCTGCGAGATCTGGCCGTGGCTGTGGAACCAGTCGTCTTCTCCCGAATGGA
GACCAAGCTCATCACGTGGGGGGCAGATACCGCCGCGTGCGGTGACATCATC
AACGGCTTGCCCGTCTCTGCCCGTAGGGGCCAGGAGATACTGCTTGGGCCAGC
CGACGGAATGGTCTCCAAGGGGTGGAGGTTGCTGGCGCCCATCACGGCGTAC
GCCCAGCAGACGAGAGGCCTCCTAGGGTGTATAATCACCAGCCTGACTGGCCG
GGACAAAAACCAAGTGGAGGGTGAGGTCCAGATCGTGTCAACTGCTACCCAAA
CCTTCCTGGCAACGTGCATCAATGGGGTATGCTGGACTGTCTACCACGGGGCC
GGAACGAGGACCATCGCATCACCCAAGGGTCCTGTCATCCAGATGTATACCAA
TGTGGACCAAGACCTTGTGGGCTGGCCCGCTCCTCAAGGTTCCCGCTCATTGA
CACCCTGCACCTGCGGCTCCTCGGACCTTTACCTGGTCACGAGGCACGCCGAT
GTCATTCCCGTGCGCCGGCGAGGTGATAGCAGGGGTAGCCTGCTTTCGCCCCG
GCCCATTTCCTACTTGAAAGGCTCCTCGGGGGGTCCGCTGTTGTGCCCCGCGG
GACACGCCGTGGGCCTATTCAGGGCCGCGGTGTGCACCCGTGGAGTGGCTAA
GGCGGTGGACTTTATCCCTGTGGAGAACCTAGAGACAACCATGAGATCCCCGG
TGTTCACGGACAACTCCTCTCCACCAGCAGTGCCCCAGAGCTTCCAGGTGGCC
CACCTGCATGCTCCCACCGGCAGCGGTAAGAGCACCAAGGTCCCGGCTGCGTA
CGCAGCCCAGGGCTACAAGGTGTTGGTGCTCAACCCCTCTGTTGCTGCAACGC
TGGGCTTTGGTGCTTACATGTCCAAGGCCCATGGGGTTGATCCTAATATCAGGA
CCGGGGTGAGAACAATTACCACTGGCAGCCCCATCACGTACTCCACCTACGGC
AAGTTCCTTGCCGACGGCGGGTGCTCAGGAGGTGCTTATGACATAATAATTTGT
GACGAGTGCCACTCCACGGATGCCACATCCATCTTGGGCATCGGCACTGTCCT
TGACCAAGCAGAGACTGCGGGGGCGAGACTGGTTGTGCTCGCCACTGCTACC
CCTCCGGGCTCCGTCACTGTGTCCCATCCTAACATCGAGGAGGTTGCTCTGTCC
ACCACCGGAGAGATCCCCTTTTACGGCAAGGCTATCCCCCTCGAGGTGATCAA
GGGGGGAAGACATCTCATCTTCTGCCACTCAAAGAAGAAGTGCGACGAGCTCG
CCGCGAAGCTGGTCGCATTGGGCATCAATGCCGTGGCCTACTACCGCGGTCTT
GACGTGTCTGTCATCCCGACCAGCGGCGATGTTGTCGTCGTGTCGACCGATGC
TCTCATGACTGGCTTTACCGGCGACTTCGACTCTGTGATAGACTGCAACACGTG
TGTCACTCAGACAGTCGATTTCAGCCTTGACCCTACCTTTACCATTGAGACAAC
CACGCTCCCCCAGGATGCTGTCTCCAGGACTCAACGCCGGGGCAGGACTGGC
AGGGGGAAGCCAGGCATCTACAGATTTGTGGCACCGGGGGAGCGCCCCTCCG
GCATGTTCGACTCGTCCGTCCTCTGTGAGTGCTATGACGCGGGCTGTGCTTGG
TATGAGCTCACGCCCGCCGAGACTACAGTTAGGCTACGAGCGTACATGAACAC
CCCGGGGCTTCCCGTGTGCCAGGACCATCTTGAATTTTGGGAGGGCGTCTTTA
CGGGCCTCACTCATATAGATGCCCACTTTCTATCCCAGACAAAGCAGAGTGGG
GAGAACTTTCCTTACCTGGTAGCGTACCAAGCCACCGTGTGCGCTAGGGCTCA
AGCCCCTCCCCCATCGTGGGACCAGATGTGGAAGTGTTTGATCCGCCTTAAAC
CCACCCTCCATGGGCCAACACCCCTGCTATACAGACTGGGCGCTGTTCAGAAT
GAAGTCACCCTGACGCACCCAATCACCAAATACATCATGACATGCATGTCGGCC
GACCTGGAGGTCGTCACGAGCACCTGGGTGCTCGTTGGCGGCGTCCTGGCTG
CTCTGGCCGCGTATTGCCTGTCAACAGGCTGCGTGGTCATAGTGGGCAGGATT
GTCTTGTCCGGGAAGCCGGCAATTATACCTGACAGGGAGGTTCTCTACCAGGA
GTTCGATGAGATGGAAGAGTGCTCTCAGCACTTACCGTACATCGAGCAAGGGA
TGATGCTCGCTGAGCAGTTCAAGCAGAAGGCCCTCGGCCTCCTGCAGACCGCG

FIGURE 26-2

TCCCGCCAAGCAGAGGTTATCACCCCTGCTGTCCAGACCAACTGGCAGAAACT
CGAGGTCTTCTGGGCGAAGCACATGTGGAATTTCATCAGTGGGATACAATACTT
GGCGGGCCTGTCAACGCTGCCTGGTAACCCCGCCATTGCTTCATTGATGGCTTT
TACAGCTGCCGTCACCAGCCCACTAACCACTGGCCAAACCCTCCTCTTCAACAT
ATTGGGGGGGTGGGTGGCTGCCCAGCTCGCCGCCCCCGGTGCCGCTACCGCC
TTTGTGGGCGCTGGCTTAGCTGGCGCCGCCATCGGCAGCGTTGGACTGGGGA
AGGTCCTCGTGGACATTCTTGCAGGGTATGGCGCGGGCGTGGCGGGAGCTCT
TGTAGCCTTCAAGATCATGAGCGGTGAGGTCCCCTCCACGGAGGACCTGGTCA
ATCTGCTGCCCGCCATCCTCTCGCCTGGAGCCCTTGTAGTCGGTGTGGTCTGC
GCAGCAATACTGCGCCGGCACGTTGGCCCGGGCGAGGGGGCAGTGCAATGGA
TGAACCGGCTAATAGCCTTCGCCTCCCGGGGGAACCATGTTTCCCCCACGCAC
TACGTGCCGGAGAGCGATGCAGCCGCCCGCGTCACTGCCATACTCAGCAGCCT
CACTGTAACCCAGCTCCTGATcgCTAGaccatggggtaccgagCGTTACTGGCCGAAGCC
GCTTGGAATAAGGCCGGTGTGCGTTTGTCTATATGTTATTTTCCACCATATTGCC
GTCTTTTGGCAATGTGAGGGCCCGGAAACCTGGCCCTGTCTTCTTGACGAGCA
TTCCTAGGGGTCTTTCCCCTCTCGCCAAAGGAATGCAAGGTCTGTTGAATGTCG
TGAAGGAAGCAGTTCCTCTGGAAGCTTCTTGAAGACAAACAACGTCTGTAGCG
ACCCTTTGCAGGCAGCGGAACCCCCCACCTGGCGACAGGTGCCTCTGCGGCCA
AAAGCCACGTGTATAAGATACACCTGCAAAGGCGGCACAACCCCAGTGCCACG
TTGTGAGTTGGATAGTTGTGGAAAGAGTCAAATGGCTCTCCTCAAGCGTATTCA
ACAAGGGGCTGAAGGATGCCCAGAAGGTACCCCATTGTATGGGATCTGATCTG
GGGCCTCGGTGCACATGCTTTACATGTGTTTAGTCGAGGTTAAAAAACGTCTAG
GCCCCCCGAACCACGGGGACGTGGTTTTCCTTTGAAAAACACGATGATAATAT
GGAGTTGATCACAAATGAACTTTTATACAAAACATACAAACAAAAACCCGTCGG
GGTGGAGGAACCTGTTTATGATCAGGCAGGTGATCCCTTATTTGGTGAAAGGG
GAGCAGTCCACCCTCAATCGACGCTAAAGCTCCCACACAAGAGAGGGGAACGC
GATGTTCCAACCAACTTGGCATCCTTACCAAAAAGAGGTGACTGCAGGTCGGG
TAATAGCAGAGGACCTGTGAGCGGGATCTACCTGAAGCCAGGGCCACTATTTT
ACCAGGACTATAAAGGTCCCGTCTATCACAGGGCCCCGCTGGAGCTCTTTGAG
GAGGGATCCATGTGTGAAACGACTAAACGGATAGGGAGAGTAACTGGAAGTG
ACGGAAAGCTGTACCACATTTATGTGTGTATAGATGGATGTATAATAATAAAAA
GTGCCACGAGAAGTTACCAAAGGGTGTTCAGGTGGGTCCATAATAGGCTTGAC
TGCCCTCTATGGGTCACAAGTTGCTCAGACACGAAAGAAGAGGGAGCAACAaag
cttGCATTGTTGGCGTGGGCAATAATAGCTATAGTTTTGTTTCAAGTTACAATGGG
AGAAAACATAACACAGTGGAACctgcagTGGTTTGACCTGGAGGTGACTGACCAT
CACCGGGATTACTTCGCTGAGTCCATATTAGTGGTGGTAGTAGCCCTCTTGGGT
GGCAGATATGTACTTTGGTTACTGGTTACATACATGGTCTTATCAGAACAGAAG
GCCTTAGGGATTCAGTATGGATCAGGGGAAGTGGTGATGATGGGCAACTTGCT
AACCCATAACAATATTGAAGTGGTGACATACTTCTTGCTGCTGTACCTACTGCT
GAGGGAGGAGAGCGTAAAGAAGTGGGTCTTACTCTTATACCACATCTTAGTGG
TACACCCAATCAAATCTGTAATTGTGATCCTACTGATGATTGGGGATGTGGTAA
AGGCCGATTCAGGGGGCCAAGAGTACTTGGGGAAAATAGACCTCTGTTTTACA
ACAGTAGTACTAATCGTCATAGGTTTAATCATAGCTAGGCGTGACCCAACTATA
GTGCCACTGGTAACAATAATGGCAGCACTGAGGGTCACTGAACTGACCCACCA
GCCTGGAGTTGACATCGCTGTGGCGGTCATGACTATAACCCTACTGATGGTTA
GCTATGTGACAGATTATTTTAGATATAAAAAATGGTTACAGTGCATTCTCAGCCT
GGTATCTGGGGTGTTCTTGATAAGAAGCCTAATATACCTAGGTAGAATCGAGAT
GCCAGAGGTAACTATCCCAAACTGGAGACCACTAACTTTAATACTATTATATTTG
ATCTCAACAACAATTGTAACGAGGTGGAAGGTTGACGTGGCTGGCCTATTGTT
GCAATGTGTGCCTATCTTATTGCTGGTCACAACCTTGTGGGCCGACTTCTTAAC
CCTAATACTGATCCTGCCTACCTATGAATTGGTTAAATTATACTATCTGAAAACT
GTTAGGACTGATATAGAAAGAAGTTGGCTAGGGGGGATAGACTATACAAGAGT
TGACTCCATCTACGACGTTGATGAGAGTGGAGAGGGCGTATATCTTTTTCCATC
AAGGCAGAAAGCACAGGGGAATTTTTCTATACTCTTGCCCCTTATCAAAGCAAC

FIGURE 26-3

ACTGATAAGTTGCGTCAGCAGTAAATGGCAGCTAATATACATGAGTTACTTAAC
TTTGGACTTTATGTACTACATGCACAGGAAAGTTATAGAAGAGATCTCAGGAGG
TACCAACATAATATCCAGGTTAGTGGCAGCACTCATAGAGCTGAACTGGTCCAT
GGAAGAAGAGGAGAGCAAAGGCTTAAAGAAGTTTTATCTATTGTCTGGAAGGT
TGAGAAACCTAATAATAAAACATAAGGTAAGGAATGAGACCGTGGCTTCTTGGT
ACGGGGAGGAGGAAGTCTACGGTATGCCAAAGATCATGACTATAATCAAGGCC
AGTACACTGAGTAAGAGCAGGCACTGCATAATATGCACTGTATGTGAGGGCCG
AGAGTGGAAAGGTGGCACCTGCCCAAAATGTGGACGCCATGGGAAGCCGATA
ACGTGTGGGATGTCGCTAGCAGATTTTGAAGAAAGACACTATAAAAGAATCTTT
ATAAGGGAAGGCAACTTTGAGGGTATGTGCAGCCGATGCCAGGGAAAGCATA
GGAGGTTTGAAATGGACCGGGAACCTAAGAGTGCCAGATACTGTGCTGAGTGT
AATAGGCTGCATCCTGCTGAGGAAGGTGACTTTTGGGCAGAGTCGAGCATGTT
GGGCCTCAAAATCACCTACTTTGCGCTGATGGATGGAAAGGTGTATGATATCAC
AGAGTGGGCTGGATGCCAGCGTGTGGGAATCTCCCCAGATACCCACAGAGTCC
CTTGTCACATCTCATTTGGTTCACGGATGCCTTTCAGGCAGGAATACAATGGCT
TTGTACAATATACCGCTAGGGGGCAACTATTTCTGAGAAACTTGCCCGTACTGG
CAACTAAAGTAAAAATGCTCATGGTAGGCAACCTTGGAGAAGAAATTGGTAATC
TGGAACATCTTGGGTGGATCCTAAGGGGGCCTGCCGTGTGTAAGAAGATCACA
GAGCACGAAAAATGCCACATTAATATACTGGATAAACTAACCGCATTTTTCGGG
ATCATGCCAAGGGGGACTACACCCAGAGCCCCGGTGAGGTTCCCTACGAGCTT
ACTAAAAGTGAGGAGGGGTCTGGAGACTGGCTGGGCTTACACACACCAAGGC
GGGATAAGTTCAGTCGACCATGTAACCGCCGGAAAAGATCTACTGGTCTGTGA
CAGCATGGGACGAACTAGAGTGGTTTGCCAAAGCAACAACAGGTTGACCGATG
AGACAGAGTATGGCGTCAAGACTGACTCAGGGTGCCCAGACGGTGCCAGATG
TTATGTGTTAAATCCAGAGGCCGTTAACATATCAGGATCCAAAGGGGCAGTCGT
TCACCTCCAAAAGACAGGTGGAGAATTCACGTGTGTCACCGCATCAGGCACAC
CGGCTTTCTTCGACCTAAAAAACTTGAAAGGATGGTCAGGCTTGCCTATATTTG
AAGCCTCCAGCGGGAGGGTGGTTGGCAGAGTCAAAGTAGGGAAGAATGAAGA
GTCTAAACCTACAAAAATAATGAGTGGAATCCAGACCGTCTCAAAAAACACAGC
AGACCTGACCGAGATGGTCAAGAAGATAACCAGCATGAACAGGGGAGACTTCA
AGCAGATTACTTTGGCAACAGGGGCAGGCAAAACCACAGAACTCCCAAAAGCA
GTTATAGAGGAGATAGGAAGACACAAGAGAGTATTAGTTCTTATACCATTAAGG
GCAGCGGCAGAGTCAGTCTACCAGTATATGAGATTGAAACACCCAAGCATCTC
TTTTAACCTAAGGATAGGGGACATGAAAGAGGGGGACATGGCAACCGGGATA
ACCTATGCATCATACGGGTACTTCTGCCAAATGCCTCAACCAAAGCTCAGAGCT
GCTATGGTAGAATACTCATACATATTCTTAGATGAATACCATTGTGCCACTCCTG
AACAACTGGCAATTATCGGGAAGATCCACAGATTTTCAGAGAGTATAAGGGTT
GTCGCCATGACTGCCACGCCAGCAGGGTCGGTGACCACAACAGGTCAAAAGC
ACCCAATAGAGGAATTCATAGCCCCCGAGGTAATGAAAGGGGAGGATCTTGGT
AGTCAGTTCCTTGATATAGCAGGGTTAAAAATACCAGTGGATGAGATGAAAGG
CAATATGTTGGTTTTTGTACCAACGAGAAACATGGCAGTAGAGGTAGCAAAGA
AGCTAAAAGCTAAGGGCTATAACTCTGGATACTATTACAGTGGAGAGGATCCA
GCCAATCTGAGAGTTGTGACATCACAATCCCCCTATGTAATCGTGGCTACAAAT
GCTATTGAATCAGGAGTGACACTACCAGATTTGGACACGGTTATAGACACGGG
GTTGAAATGTGAAAAGAGGGTGAGGGTATCATCAAAGATACCCTTCATCGTAA
CAGGCCTTAAGAGGATGGCCGTGACTGTGGGTGAGCAGGCGCAGCGTAGGGG
CAGAGTAGGTAGAGTGAAACCCGGGAGGTATTATAGGAGCCAGGAAACAGCA
ACAGGGTCAAAGGACTACCACTATGACCTCTTGCAGGCACAAAGATACGGGAT
TGAGGATGGAATCAACGTGACGAAATCCTTTAGGGAGATGAATTACGATTGGA
GCCTATACGAGGAGGACAGCCTACTAATAACCCAGCTGGAAATACTAAATAATC
TACTCATCTCAGAAGACTTGCCAGCCGCTGTTAAGAACATAATGGCCAGGACTG
ATCACCCAGAGCCAATCCAACTTGCATACAACAGCTATGAAGTCCAGGTCCCG
GTCCTGTTCCCAAAAATAAGGAATGGAGAAGTCACAGACACCTACGAAAATTAC
TCGTTTCTAAATGCCAGAAAGTTAGGGGAGGATGTGCCCGTGTATATCTACGCT

FIGURE 26-4

ACTGAAGATGAGGATCTGGCAGTTGACCTCTTAGGGCTAGACTGGCCTGATCC
TGGGAACCAGCAGGTAGTGGAGACTGGTAAAGCACTGAAGCAAGTGACCGGG
TTGTCCTCGGCTGAAAATGCCCTACTAGTGGCTTTATTTGGGTATGTGGGTTAC
CAGGCTCTCTCAAAGAGGCATGTCCCAATGATAACAGACATATATACCATCGAG
GACCAGAGACTAGAAGACACCACCCACCTCCAGTATGCACCCAACGCCATAAA
AACCGATGGGACAGAGACTGAACTGAAAGAACTGGCGTCGGGTGACGTGGAA
AAAATCATGGGAGCCATTTCAGATTATGCAGCTGGGGGACTGGAGTTTGTTAA
ATCCCAAGCAGAAAAGATAAAAACAGCTCCTTTGTTTAAAGAAAACGCAGAAGC
CGCAAAAGGGTATGTCCAAAAATTCATTGACTCATTAATTGAAAATAAAGAAGA
AATAATCAGATATGGTTTGTGGGGAACACACACAGCACTATACAAAAGCATAGC
TGCAAGACTGGGGCATGAAACAGCGTTTGCCACACTAGTGTTAAAGTGGCTAG
CTTTTGGAGGGGAATCAGTGTCAGACCACGTCAAGCAGGCGGCAGTTGATTTA
GTGGTCTATTATGTGATGAATAAGCCTTCCTTCCCAGGTGACTCCGAGACACAG
CAAGAAGGGAGGCGATTCGTCGCAAGCCTGTTCATCTCCGCACTGGCAACCTA
CACATACAAAACTTGGAATTACCACAATCTCTCTAAAGTGGTGGAACCAGCCCT
GGCTTACCTCCCCTATGCTACCAGCGCATTAAAAATGTTCACCCCAACGCGGCT
GGAGAGCGTGGTGATACTGAGCACCACGATATATAAAACATACCTCTCTATAAG
GAAGGGGAAGAGTGATGGATTGCTGGGTACGGGGATAAGTGCAGCCATGGAA
ATCCTGTCACAAAACCCAGTATCGGTAGGTATATCTGTGATGTTGGGGGTAGG
GGCAATCGCTGCGCACAACGCTATTGAGTCCAGTGAACAGAAAAGGACCCTAC
TTATGAAGGTGTTTGTAAAGAACTTCTTGGATCAGGCTGCAACAGATGAGCTGG
TAAAAGAAAACCCAGAAAAAATTATAATGGCCTTATTTGAAGCAGTCCAGACAA
TTGGTAACCCCCTGAGACTAATATACCACCTGTATGGGGTTTACTACAAAGGTT
GGGAGGCCAAGGAACTATCTGAGAGGACAGCAGGCAGAAACTTATTCACATTG
ATAATGTTTGAAGCCTTCGAGTTATTAGGGATGGACTCACAAGGGAAAATAAG
GAACCTGTCCGGAAATTACATTTTGGATTTGATATACGGCCTACACAAGCAAAT
CAACAGAGGGCTGAAGAAAATGGTACTGGGGTGGGCCCCTGCACCCTTTAGTT
GTGACTGGACCCCTAGTGACGAGAGGATCAGATTGCCAACAGACAACTATTTG
AGGGTAGAAACCAGGTGCCCATGTGGCTATGAGATGAAAGCTTTCAAAAATGT
AGGTGGCAAACTTACCAAAGTGGAGGAGAGCGGGCCTTTCCTATGTAGAAACA
GACCTGGTAGGGGACCAGTCAACTACAGAGTCACCAAGTATTACGATGACAAC
CTCAGAGAGATAAAACCAGTAGCAAAGTTGGAAGGACAGGTAGAGCACTACTA
CAAAGGGGTCACAGCAAAAATTGACTACAGTAAAGGAAAAATGCTCTTGGCCA
CTGACAAGTGGGAGGTGGAACATGGTGTCATAACCAGGTTAGCTAAGAGATAT
ACTGGGGTCGGGTTCAATGGTGCATACTTAGGTGACGAGCCCAATCACCGTGC
TCTAGTGGAGAGGGACTGTGCAACTATAACCAAAAACACAGTACAGTTTCTAAA
AATGAAGAAGGGGTGTGCGTTCACCTATGACCTGACCATCTCCAATCTGACCA
GGCTCATCGAACTAGTACACAGGAACAATCTTGAAGAGAAGGAAATACCCACC
GCTACGGTCACCACATGGCTAGCTTACACCTTCGTGAATGAAGACGTAGGGAC
TATAAAACCAGTACTAGGAGAGAGAGTAATCCCCGACCCTGTAGTTGATATCAA
TTTACAACCAGAGGTGCAAGTGGACACGTCAGAGGTTGGGATCACAATAATTG
GAAGGGAAACCCTGATGACAACGGGAGTGACACCTGTCTTGGAAAAAGTAGA
GCCTGACGCCAGCGACAACCAAAACTCGGTGAAGATCGGGTTGGATGAGGGT
AATTACCCAGGGCCTGGAATACAGACACATACACTAACAGAAGAAATACACAA
CAGGGATGCGAGGCCCTTCATCATGATCCTGGGCTCAAGGAATTCCATATCAA
ATAGGGCAAAGACTGCTAGAAATATAAATCTGTACACAGGAAATGACCCCAGG
GAAATACGAGACTTGATGGCTGCAGGGCGCATGTTAGTAGTAGCACTGAGGGA
TGTCGACCCTGAGCTGTCTGAAATGGTCGATTTCAAGGGGACTTTTTTAGATAG
GGAGGCCCTGGAGGCTCTAAGTCTCGGGCAACCTAAACCGAAGCAGGTTACCA
AGGAAGCTGTTAGGAATTTGATAGAACAGAAAAAAGATGTGGAGATCCCTAAC
TGGTTTGCATCAGATGACCCAGTATTTCTGGAAGTGGCCTTAAAAAATGATAAG
TACTACTTAGTAGGAGATGTTGGAGAGGTAAAAGATCAAGCTAAAGCACTTGG
GGCCACGGATCAGACAAGAATTATAAAGGAGGTAGGCTCAAGGACGTATGCCA
TGAAGCTATCTAGCTGGTTCCTCAAGGCATCAAACAAACAGATGAGTTTAACTC

FIGURE 26-5

CACTGTTTGAGGAATTGTTGCTACGGTGCCCACCTGCAACTAAGAGCAATAAG
GGGCACATGGCATCAGCTTACCAATTGGCACAGGGTAACTGGGAGCCCCTCGG
TTGCGGGGTGCACCTAGGTACAATACCAGCCAGAAGGGTGAAGATACACCCAT
ATGAAGCTTACCTGAAGTTGAAAGATTTCATAGAAGAAGAAGAGAAGAAACCT
AGGGTTAAGGATACAGTAATAAGAGAGCACAACAAATGGATACTTAAAAAAAT
AAGGTTTCAAGGAAACCTCAACACCAAGAAAATGCTCAACCCTGGGAAACTATC
TGAACAGTTGGACAGGGAGGGGCGCAAGAGGAACATCTACAACCACCAGATT
GGTACTATAATGTCAAGTGCAGGCATAAGGCTGGAGAAATTGCCAATAGTGAG
GGCCCAAACCGACACCAAAACCTTTCATGAGGCAATAAGAGATAAGATAGACA
AGAGTGAAAACCGGCAAAATCCAGAATTGCACAACAAATTGTTGGAGATTTTCC
ACACGATAGCCCAACCCACCCTGAAACACACCTACGGTGAGGTGACGTGGGAG
CAACTTGAGGCGGGGATAAATAGAAAGGGGGCAGCAGGCTTCCTGGAGAAGA
AGAACATCGGAGAAGTATTGGATTCAGAAAAGCACCTGGTAGAACAATTGGTC
AGGGATCTGAAGGCCGGGAGAAAGATAAAATATTATGAAACTGCAATACCAAA
AAATGAGAAGAGAGATGTCAGTGATGACTGGCAGGCAGGGGACCTGGTGGTT
GAGAAGAGGCCAAGAGTTATCCAATACCCTGAAGCCAAGACAAGGCTAGCCAT
CACTAAGGTCATGTATAACTGGGTGAAACAGCAGCCCGTTGTGATTCCAGGAT
ATGAAGGAAAGACCCCCTTGTTCAACATCTTTGATAAAGTGAGAAAGGAATGG
GACTCGTTCAATGAGCCAGTGGCCGTAAGTTTTGACACCAAAGCCTGGGACAC
TCAAGTGACTAGTAAGGATCTGCAACTTATTGGAGAAATCCAGAAATATTACTA
TAAGAAGGAGTGGCACAAGTTCATTGACACCATCACCGACCACATGACAGAAG
TACCAGTTATAACAGCAGATGGTGAAGTATATATAAGAAATGGGCAGAGAGGG
AGCGGCCAGCCAGACACAAGTGCTGGCAACAGCATGTTAAATGTCCTGACAAT
GATGTACGCCTTCTGCGAAAGCACAGGGGTACCGTACAAGAGTTTCAACAGGG
TGGCAAGGATCCACGTCTGTGGGGATGATGGCTTCTTAATAACTGAAAAAGGG
TTAGGGCTGAAATTTGCTAACAAAGGGATGCAGATTCTTCATGAAGCAGGCAA
ACCTCAGAAGATAACGGAAGGGGAAAAGATGAAAGTTGCCTATAGATTTGAGG
ATATAGAGTTCTGTTCTCATACCCCAGTCCCTGTTAGGTGGTCCGACAACACCA
GTAGTCACATGGCCGGGAGAGACACCGCTGTGATACTATCAAAGATGGCAACA
AGATTGGATTCAAGTGGAGAGAGGGGTACCACAGCATATGAAAAAGCGGTAG
CCTTCAGTTTCTTGCTGATGTATTCCTGGAACCCGCTTGTTAGGAGGATTTGCCT
GTTGGTCCTTTCGCAACAGCCAGAGACAGACCCATCAAAACATGCCACTTATTA
TTACAAAGGTGATCCAATAGGGGCCTATAAAGATGTAATAGGTCGGAATCTAA
GTGAACTGAAGAGAACAGGCTTTGAGAAATTGGCAAATCTAAACCTAAGCCTG
TCCACGTTGGGGATCTGGACTAAGCACACAAGCAAAAGAATAATTCAGGACTG
TGTTGCCATTGGGAAAGAAGAGGGCAACTGGCTAGTTAACGCCGACAGGCTGA
TATCCAGCAAAACTGGCCACTTATACATACCTGATAAAGGCTTTACATTACAAG
GAAAGCATTATGAGCAACTGCAGCTAAGAACAGAGACAAACCCGGTCATGGGG
GTTGGGACTGAGAGATACAAGTTAGGTCCCATAGTCAATCTGCTGCTGAGAAG
GTTGAAAATTCTGCTCATGACGGCCGTCGGCGTCAGCAGCTGAgacaaaatgtatatattgt
aaataaattaatccatgtacatagtgtatataaatatagttgggaccgtccacctcaagaagacgacacgcccaacacgcacagctaaac
agtagtcaagattatctacctcaagataacactacatttaatgcacacagcactttagctgtatgaggatacgcccgacgtctatagttggac
tagggaagacctctaacagccccc

FIGURE 26-6

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US99/08850

A. CLASSIFICATION OF SUBJECT MATTER

IPC(6) :A61K 39/29, 39/295; C12Q 1/70; C12N 7/01; C07H 21/02
US CL :424/218.1, 228.1; 435/5, 235.1; 536/23.72
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 424/218.1, 228.1; 435/5, 235.1; 536/23.72

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

APS; Derwent/WEST; DIALOG

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X,P	FROLOV et al. cis-acting RNA elements required for replication of bovine viral diarrhea virus-hepatitis C virus 5' nontranslated region chimeras. RNA. November 1998, Vol. 4, pages 1418-1435, see entire document.	1-8, 10-21
Y,P	MALET et al. Yellow fever 5' noncoding region as a potential element to improve hepatitis C virus production through modification of translational control. Biochem. Biophys. Res. Commun. 18 December 1998, Vol. 253, No. 2, pages 257-264, see entire document.	1-8, 10-21

[X] Further documents are listed in the continuation of Box C. [ ] See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search

19 JULY 1999

Date of mailing of the international search report

10 SEP 1999

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INTERNATIONAL SEARCH REPORT

International application No.
PCT/US99/08850

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	LU et al. Poliovirus chimeras replicating under the translational control of genetic elements of hepatitis C virus reveal unusual properties of the internal ribosomal entry site of hepatitis C virus. Proc. Natl. Acad. Sci. USA. 20 February 1996, Vol. 93, No. 4, pages 1412-1417, see entire document.	1-8, 10-21
Y	VASSILEV et al. Authentic and chimeric full-length genomic cDNA clones of bovine viral diarrhea virus that yield infectious transcripts. J. Virol. January 1997, Vol. 71, No. 1, pages 471-478, see entire document.	1-8, 10-21
Y	VENUGOPAL et al. Towards a new generation of flavivirus vaccines. Vaccines. 1994, Vol. 12, No. 11, pages 966-975, see entire document.	1-8, 10-21

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US99/08850

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☒ Claims Nos.: 9
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

CLAIM 9 RECITES "SEQ ID NO:X" WHICH EXPRESSION IS NOT UNDERSTOOD.

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest.

☐ No protest accompanied the payment of additional search fees.

CORRECTED VERSION*

PCT

WORLD INTELLECTUAL PROPERTY ORGANIZATION
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(72) Inventors; and
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(54) Title: CHIMERAS OF HEPATITIS C VIRUS AND BOVINE VIRAL DIARRHEA VIRUS

(57) Abstract

Disclosed is a polynucleotide comprising a chimeric viral RNA which contains: a 5' nontranslated region (5' NTR), an open reading frame (ORF) region, and a 3' nontranslated region (3' NTR) wherein at least one of said regions is chimeric. The chimeric region comprises a first nucleotide sequence from a pestivirus in operable linkage with a heterologous nucleotide sequence. The chimeric viral RNA is replication–competent. Preferably the pestivirus sequence is from a bovine viral diarrhea virus and the heterologous nucleotide sequence is from a hepatitis C virus. Also disclosed are a method for identifying compounds having antiviral activity agaisnt hepatitis C virus, a genetically–engineered chimeric RNA virus and a vaccine against bovine viral diarrhea virus.

*(Referred to in PCT Gazette No. 31/2000, Section II)